THE UNIVERSITY OF ALBERTA

FUNCTIONAL ASPECTS OF SOLEUS MUSCLE SPINDLES DURING LOCOMOTION IN PREMAMMILLARY CATS

by

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(C)

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The undersigned certify that they have read, and recommend to the Faculty of Graduate Studies and Research for acceptance, a thesis entitled FUNCTIONAL ASPECTS OF SOLEUS MUSCLE SPINDLES DURING LOCOMOTION IN PREMAMMILLARY CATS submitted by Jennifer B. H. Taylor in partial fulfilment of the requirements for the degree of Master of Science.

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ABSTRACT

The purpose of this thesis was to determine the functional effectiveness of the fusimotor activity on the response of muscle spindle afferents to stretch during locomotion in the cat. Anatomically, the muscle spindle is provided with a versatile sensitivity control such that the sensitivity can be set to a desired level, or varied, quite separately from the prevalent mechanical conditions. To determine the extent to which this system is utilized during locomotion, the stretch sensitivities of soleus primary and secondary muscle spindle afferents were studied during locomotion in the premammillary cat. One hindlimb was fixed and stretches were applied to its soleus muscle. The cat walked with its other three legs on a treadmill. Although its length was controlled by a servo motor, the isolated soleus muscle of the fixed hindlimb contracted with the walking rhythm. The activity in single soleus muscle spindle afferents was recorded in dissected dorsal root filaments. The stretch responses of the afferents were compared at different times in the step cycle.

The sensitivity to stretch of primary afferents was modulated during the step cycle. It was high during the period of active force development which corresponds, in a freely walking cat, to the support phase of the step cycle. It was low in the absence of force. These changes in sensitivity would act together with the locomotor changes in the α-motoneuron membrane potential to modulate the gain of

the stretch reflex. A high reflex gain during the support phase allows the ongoing extensor activity to be reinforced, such as when changes in the terrain are encountered. A reduction to a lower gain then prevents the passive stretch of the muscle in the swing phase from producing undesirable reflex extensor activity which would oppose the movement.

The sensitivity to stretch of secondary afferents was maintained at a low, relatively tonic level throughout the step cycle. During normal walking, this would allow the secondary afferent to signal muscle length throughout the whole step cycle, despite the quite extensive excursions in muscle length which occur.

The stretch responses were also studied in anesthetized cats during controlled stimulation of γ-motoneurons. The γ-motoneurons were stimulated with tonic rates and with a modulated rate. These rates were representative of the activity patterns that we have recorded from γ-motoneurons during walking in other experiments with premammillary cats. A dynamic and static γ-motoneuron influencing the same afferent were stimulated with a phasic and tonic pattern to determine their interaction on the stretch response. From these experiments the stretch responses during walking could be interpreted in terms of the fusimotor activity influencing the spindles. The results were consistent with a tonic pattern of activity occurring in static γ-motoneurons and a modulated pattern of activity occurring in dynamic γ-motoneurons.

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CHAPTER ONE

PREAMBLE

Rhythmic movements are produced by coordinated patterns of muscle activity, alternating in a stereotypic manner. An example is locomotion in the cat. Locomotion must be controlled so that the muscles of a limb are activated in the proper sequence to give the appropriate cyclic movements of the joints, so that the movement of each limb is coordinated with the others, and so that the movements are adapted to the external conditions and the goals of the animal (Orlovsky & Shik, 1976; Grillner, 1981).

In the cat, the rhythmic walking pattern is generated in the spinal cord. Descending inputs activate and control the spinal locomotor centres. Activity in sensory fibers, from muscle, joint, and cutaneous receptors, accompanies locomotion and may also contribute to its control (Grillner, 1975; Grillner & Zangger, 1975, 1979; Perret, 1976; Shik & Orlovsky, 1976; Patla et al, 1985).

Muscle stretch receptors, the muscle spindles, are the only proprioceptors to receive their own motor innervation: from static and dynamic gamma motoneurons. Gamma (γ) motoneurons affect the bias of the muscle spindle afferents and their sensitivity to stretch. Consequently, the discharges of the primary and secondary afferents are a result of the complex interaction at the muscle spindle between length changes and γ-motoneuron activity. As a cat walks, the ankle extensor muscles change length cyclically,

and also, from environmental perturbations, sometimes unexpectedly. The locomotor γ-motoneuron activity influences the responses of the primary and secondary endings to these length changes. The flexibility in the response to length changes provided by the independent γ-motoneuron innervation of the spindles could be meaningful in the control of rhythmic movements such as locomotion (Matthews, 1981a; Hulliger, 1984).

CHAPTER TWO

PURPOSE

The aim of this study was to examine the responses of primary and secondary spindle afferents to stretch during locomotion in the premammillary cat. One hindlimb was clamped and its soleus muscle oscillated about a preset length while the activity from single soleus muscle spindle afferents was recorded. The cat walked with its three other legs on a treadmill. This preparation allowed the afferent response to controlled stretches alone to be determined at different times in the step cycle without the complicating effects of muscle length changes produced by locomotion. Attention was directed to comparison of the responses of primary and secondary afferents at different times during the step cycle, with the responses when the cat was standing quietly. So that the fusimotor activity could be interpreted from the afferent recordings, the stretch responses were also studied in anesthetized cats during controlled stimulation of identified γ-motoneurons.

Two patterns of activity in the γ-motoneurons from ankle extensor muscles have previously been observed during locomotion in the premammillary cat (Murphy et al, 1984). Some fusimotor axons (γ_p) showed a deeply modulated rate of firing during the step cycle; the others (γ_t) showed a high steady firing rate. It was suggested that the γ_p and γ_t axons correspond to dynamic and static fusimotor neurons, respectively.

An intention of the analysis of the afferent responses during walking was to determine the functional effectiveness of the fusimotor activity. A primary ending can be influenced by a number of dynamic and static γ-motoneurons. Static and dynamic γ-motoneurons have quite different effects on the stretch sensitivity of a primary ending (Hulliger et al, 1977a; Matthews, 1981a). Thus the net effect of the combined fusimotor actions is difficult to predict (Hulliger et al, 1977b). Afferent recordings are required to determine how the activity of γ- motoneurons during locomotion influences the response to length changes.

Another objective was to more conclusively identify the activity patterns in static and dynamic γ-motoneurons on the basis of the stretch responses of the primary and secondary afferents during the step cycle. This should be possible since only static γ-motoneurons significantly affect secondary endings (Hulliger, 1984).

The stretch reflex is centrally modulated during the step cycle (Akazawa et al, 1982) through phasic changes in the alpha (α) motoneuron membrane potential (Shefchyk et al, 1984). A third purpose of the study was to determine whether variations in afferent input contribute to modulation of the reflex.

CHAPTER THREE

INTRODUCTION

3.1 LOCOMOTION

3.1.1 DESCRIPTION

Much of our knowledge of cat locomotion has come from studies of the hindlimb muscles. The step cycle of the hindlimb can be described in terms of one flexion phase, F, and three extension phases, E1, E2, and E3 (Engberg & Lundberg, 1969; Grillner, 1975). After the foot leaves the ground, all the joints of the limb flex and the foot is brought up and forward. This first part of the swing phase is F. E1 commences as the knee and ankle joints extend while the limb still travels forward in the swing phase. Activity in the hip extensors at this time decelerates the limb. Knee extension increases the stride length. Ankle extensor activity begins just before footfall, an event which marks the end of E1. The ankle extension of E1 lowers the paw slightly to bring the footpads in contact with the ground and allow digitigrade support. In the first part of the support phase, E2, the limb yields under the weight of the body, flexing the ankle and knee joints and stretching the active ankle extensors. All the extensors are strongly active in this phase; the activity continues until just before the foot leaves the ground. In E3, all the joints extend as the weight travels forward over the limb and the extensor muscles slowly shorten a small amount. After the

foot leaves the ground the limb extension is briefly continued until the flexor activity is sufficient to overcome the backward momentum. Activity in the ankle and knee flexors generally occurs in three bursts: at the end of E3, at the start of F, and towards the end of E1. Hip flexor activity starts later and ends later than the activity in the ankle and knee flexors (Engberg & Lundberg, 1969).

The major proximal hindlimb muscles are bifunctional and produce opposite movements at the hip and knee joints. Since ankle flexors and extensors show simpler activity patterns during locomotion, they have received more attention.

3.1.2 CENTRAL GENERATION

The patterns of locomotion can be produced in the spinal cord, even without cyclic sensory feedback. Brown, in 1911 (Wetzel & Stuart, 1976), first reported stepping movements of the de-afferented hindlimbs following transection of the cat's spinal cord. Many years later, rhythmic reciprocal activity in flexors and extensors of de-afferented hindlimbs was observed in chronic spinal kittens and in acute spinal cats administered Clonidine or Dopa and with tonic stimulation of the L7 or L6 dorsal root (Grillner & Zangger, 1974, 1979). Alternating bursts of activity in flexor and extensor motoneurons have been recorded, during tonic stimulation of a dorsal root, in curarized acute spinal cats (Grillner & Zangger, 1979).

That phasic afferent input is not necessary for the generation of a co-ordinated stepping behaviour has also been supported by experiments in de-afferented mesencephalic cats during tonic stimulation of the mesencephalic locomotor region (Grillner & Zangger, 1975) and in de-afferented or curarized decorticate cats (Perret, 1976). Not only was there alternation of flexor and extensor activity but the timing of the electromyogram (EMG) bursts, including the initiation of the extensor activity before footfall, did not change following de-afferentation in the mesencephalic cats.

3.1.3 PERIPHERAL CONTROL

The control of locomotion does rely on sensory feedback. Without it there can be considerable step to step variability in the timing of the EMG bursts and co-ordination of the limbs may be periodically lost (Grillner, 1981). The force of the step may be decreased (Wetzel et al, 1976). The animal can not adapt to changes in the environmental conditions, such as treadmill speed.

Sensory feedback allows adjustment of the centrally programmed movements, which would be advantageous when the progress of a movement is altered or unexpected changes in muscle length or load occur. Two mechanisms, one dependent on hip position, the other on muscle load, control the stance phase so that it is appropriate for the external conditions (Grillner, 1979; Andersson et al, 1981).

If one limb of a stepping spinal cat was stopped in

early or mid extension, the extensor activity in that limb was maintained (Grillner, 1975, 1981; Grillner & Rossignol, 1978). During fictive locomotion, in which muscle contraction is prevented by the action of curare, the frequency of the alternating activity of the alpha motoneurons could be decreased by slower cyclic passive movements of the hindlimb. Joint or muscle afferents from the hip were responsible for this effect (Andersson et al, 1981). Thus during stepping, if the hip was extended more slowly than expected, afferent signals would prevent the extensor activity from ending too soon, so that support would be continued (Andersson et al, 1981). In the stepping cat, extensor activity ended and flexor activity began when the limb was stopped in a position caudal to mid E3 (Grillner, 1975, 1981; Grillner & Rossignol, 1978). The burst frequency during fictive locomotion could be entrained to a faster cyclic movement of the hindlimb (Andersson et al, 1981). Thus, if a limb was extended rapidly so that it was no longer supporting the animal, flexion could ensue (Andersson et al, 1981).

If an ankle extensor was stretched during the stance phase of locomotion in the thalamic cat, extensor activity was maintained and flexor activity inhibited (Pearson & Duysens, 1976). Stimulation of the ventral root or of the muscle directly, which caused an increase in tension without a change in length, produced the same response as the muscle stretch. Flexion will thus not be initiated during locomotion until the extensors are unloaded. Normally, the

load on the extensors decreases as the body passes forward over the limb, so that by the end of the support phase the load is diminished sufficiently so as not to prevent flexion. Since a perturbation that causes slow extension of the limb also causes the load to be maintained for a longer period, the control mechanisms dependent on hip position and load evidently work co-operatively to ensure that flexion occurs at the appropriate time (Grillner, 1979). The regulation of the stance phase by these mechanisms may be important when the cat changes speed, changes direction or circles, or when an uneven or sloping terrain is unexpectedly encountered (Andersson et al, 1981).

3.1.4 REFLEX MODULATION

During locomotion, some reflex responses are dependent on the phase of the step cycle in which the stimulation is applied. The extensor thrust reflex is only effective in the stance phase. In the thalamic cat, stimulation of the pad, plantar surface or sural nerve resulted in increased activity in the ipsilateral ankle extensor, and a delay in the onset of the flexor activity (and contralateral extensor activity) when the stimulation was applied during the support phase. No response was elicited when the stimulation was applied during the swing phase: the time of flexor activity (Duysens & Pearson, 1976).

The stretch reflex in ankle extensors is also modulated during the step cycle. In the mesencephalic cat, the change in amplitude of the reflex response approximately followed

the time course of the locomotor muscle activity (Akazawa et al, 1982). Changes in muscle spindle sensitivity could contribute to the stretch reflex modulation and this possibility is examined in this thesis.

Electrical stimulation of the muscle nerve at a strength sufficient to excite only the Group I afferents resulted in modulation of the H-reflex during the step cycle similar to that seen for the stretch reflex (Akazawa et al, 1982). Since the afferent input was constant under this condition, central factors must have accounted for the H-reflex modulation and would also contribute to the stretch reflex modulation. One central gating mechanism is the cyclic change in the excitability of the alpha motoneurons, i.e. in the membrane potential, so that the probability of an EPSP of constant size reaching threshold also varies cyclically (Grillner, 1981; Akazawa et al, 1982). A second potential mechanism is presynaptic modulation of afferent activity so that the size of the EPSP varies cyclically (Grillner, 1981; Akazawa et al, 1982). Since little change in the amplitude of the Ia EPSP's was observed by recording intracellularly from α-motoneurons, importance was attributed to the former mechanism (Shefchyk et al, 1984). The possibility that changes in muscle spindle sensitivity, due to varying fusimotor activity, contributed to the stretch reflex modulation could not, of course, be excluded.

The reaction to tactile stimulation of the dorsum of the paw during locomotion is another example of a phase

dependent reflex (Forssberg, 1979). Stimulation applied during the swing phase of locomotion in spinal and intact cats evoked increased ipsilateral flexor activity, so that the foot would be lifted clear of the offending object. Stimulation applied during the stance phase caused an increase in the ongoing extensor activity in spinal cats, thrusting the foot backward, followed by enhanced flexor activity during the succeeding swing phase. In intact cats, inhibitory and excitatory effects during extension balanced so that the support phase did not change much. In addition to the phasic variation in motoneuron excitability, premotoneuronal modulation of transmission in the reflex pathway contributes to the phase dependency of this stumbling corrective reaction (Andersson et al, 1978; Forssberg, 1979; Grillner, 1981).

3.2 THE MUSCLE SPINDLE

3.2.1 STRUCTURE

As early as 1900, based mainly on descriptions by Sherrington and Ruffini, muscle spindles have been recognized as sense organs that are under motor control. In recent years especially, the evolving view of spindle structure has become increasingly complex (Boyd, 1981a).

The soleus muscle in the cat contains about fifty muscle spindles (Prochazka, 1981), each of which is approximately 1 cm in length. The fusiform shape of the muscle spindle is due to a centrally expanded connective

tissue capsule. Intrafusal muscle fibers, in parallel, extend beyond the capsular sleeve (Boyd, 1980). A muscle spindle contains one, or occasionally two, dynamic nuclear bag (bag_1) fibers, one static nuclear bag (bag_2) fiber, and three to five chain fibers (Boyd, 1981a).

A bag_1 fiber is innervated by one or two dynamic γ-motoneurons. The bag_2 fiber and chain fibers of a spindle are innervated by three to seven static γ-motoneurons (Boyd, 1980). A static γ-motoneuron can supply just chain fibers, just the bag_2 fiber, or both types, in a particular spindle (Laporte & Emonet-Denand, 1973; Boyd, 1981a; Barker, 1983). At least in tenuissimus, in all the spindles it enters, a static γ-motoneuron is consistent as to the type of intrafusal fiber it supplies, even though it may supply the other type in addition (Boyd, 1983; Boyd et al, 1983). On the basis of this predominantly selective activation, the possibility that there could be two functional classes of static γ-motoneuron, has recently been proposed (Boyd et al, 1983; Hulliger, 1984). Following a long controversy, it is now generally believed that static γ-motoneurons do not supply bag_1 fibers (Boyd, 1981a).

In addition to their control by γ-motoneurons, muscle spindles may be innervated by beta (β) motoneurons. A dynamic β-axon supplies the bag_1 fiber and slow twitch extrafusal fibers. A static β-axon supplies the longest chain fiber and fast twitch extrafusal fibers (Matthews, 1981b). The incidence of skeleto-fusimotor innervation of spindles in soleus is thought to be very low (Prochazka,

1981).

Myofilaments are scarce in the central region of the intrafusal fibers, particularly of the bag fibers in which the nuclei are densely packed (Matthews, 1981a). The sensory terminals are located on this part of the fiber. A spindle, on average, is innervated by one primary and one secondary afferent, although the number of secondary afferents is variable: 0 to 4. (Branches from one primary afferent form spiral endings about the equatorial region of every intrafusal fiber in the spindle. Branches from a secondary afferent form similar spiral endings, juxtaequatorially, around the chain fibers (Boyd, 1980). Flower spray endings from secondary afferents have been found on the bag_1 and bag_2 fibers, but the relative contact area of the secondary endings on these fibers was 10% and 20%, respectively (Banks et al, 1981; Barker, 1983).

3.2.2 RESPONSES OF PRIMARY AND SECONDARY AFFERENTS TO STRETCH

Stretch of the muscle extends the sensory endings, causing the afferent firing rate to increase. The functional distinctiveness of the two types of spindle afferents, classified on the basis of conduction velocity, was originally discovered less than 25 years ago (Matthews, 1981a). Since then, the stretch responses of the primary and secondary spindle afferents have been well described (reviewed by Matthews, 1972, 1973, 1981b).

Primary afferents are much more responsive to the

occurrence of movement. They have greater dynamic sensitivity to large ramp and triangular stretches, a lower threshold for tendon taps, and can be selectively activated by vibration (Matthews, 1972, 1981b). The dynamic index (the decrease in firing rate, over 0.5 s, following the completion of the constant velocity phase of a stretch) has been commonly used to assess the dynamic responsiveness of the afferent to large ramp stretches. It is much greater for primary than secondary afferents: for 10 mm/s ramp stretches by a factor of 6 (Matthews, 1972). At the initiation of a constant velocity stretch, primary afferents often exhibit a rapid increase in firing rate; secondary afferents do not. Another difference between the passive endings (when the spindle is not influenced by any fusimotor activity) is that primary afferents fall silent on the release phase of a ramp or triangular stretch, whereas secondary afferents show a progressive decrease in firing rate (Matthews, 1972, 1973). Primary and secondary afferents are quite similar in terms of static position sensitivity (change in firing rate / change in muscle length) measured after completion of the adaptation to the stretch (Matthews, 1972) or corrected for adaptation (Boyd, 1981b), although the absolute firing rate may be slightly higher for secondary afferents (Hulliger, 1984).

A muscle spindle afferent responds to stretches over a wide range of physiological amplitudes in a nonlinear manner. An afferent is much more responsive to stretches of

small amplitude than to large stretches (Matthews, 1981a). A linear range does exist for the small amplitude stretches to which the afferent is highly sensitive (Matthews & Stein, 1969). Within this range, the primary afferent is about 5 to 10 times more sensitive than the secondary afferent (Cussons et al, 1977). However, there is no large difference between primary and secondary endings in the relative sensitivity to length and velocity for small amplitude stretches at physiologically important frequencies, as determined by frequency response analysis (Matthews & Stein, 1969). The linear range for primary afferents is up to about 0.1 mm for 1 Hz sinusoidal stretches and decreases with increases in frequency (Hulliger, 1984). The sensitivity of the primary afferent to small stretches within the linear range is 10 to 100 times the sensitivity to large amplitude stretches (Matthews, 1981b). The constant high sensitivity to small stretches, and the progressive decrease in sensitivity with increases in amplitude outside the linear range, is known as gain compression nonlinearity (Hulliger, 1984). Functionally, it allows the primary afferent to adequately signal the occurrence of small movements and not be saturated during the progression of large movements (Matthews, 1981b).

The reason for the quite different sensitivity to large and small amplitude stretches is due to the existence of cross-bridges at the poles of the intrafusal muscle fibers (Hulliger et al, 1977a; Matthews, 1981b). The stiffness of

the attached cross-bridges results in small stretches being transmitted mainly to the central sensorial region. During large stretches, however, the cross-bridges break and reform so that part of the length change is taken up in the polar regions. Following a large stretch, the high sensitivity to small stretches is reset (Matthews, 1981b). During a slow gradual stretch, the high sensitivity to small stretches is not maintained (Hulliger, 1984).

3.2.3 STATIC AND DYNAMIC FUSIMOTOR ACTIONS

Contraction of the intrafusal fibers extends the sensory endings and affects the response to muscle length changes. This has been investigated by the technique of stimulating single γ-motoneurons in the ventral roots and recording from single afferents affected by the particular γ-motoneuron, while applying muscle stretches (Boyd, 1980).

Under static conditions, repetitive stimulation of a dynamic γ-motoneuron (γ_D) causes a small increase in the primary afferent rate (Matthews, 1972; Boyd, 1980, 1983). A static γ-motoneuron (γ_S) has a stronger direct excitatory effect on spindle activity (Matthews, 1972, 1981a; Boyd, 1980, 1983; Hulliger, 1984). It may drive the primary afferent; i.e., cause the afferent to fire once for each stimulation pulse (Boyd, 1980, 1983). The firing rate of a secondary afferent is increased by static γ-motoneuron stimulation. The static position sensitivity of the secondary afferent is also often increased, but it may be decreased or not altered at all (Boyd, 1980, 1983; Jami &

Petit, 1981; Hulliger, 1984).

During ramp stretches, the velocity dependent response of the primary afferent, assessed by the dynamic index, is increased by γ_D stimulation (Crowe & Matthews, 1964; Matthews, 1972, 1981b) but is usually decreased by γ_S stimulation (Brown et al, 1965; Matthews, 1981b; Hulliger, 1984). Stimulation of a γ_D also markedly increases the length sensitivity of the primary afferent measured during the ramp movement (dynamic position sensitivity) much more than any effect it may have on the static position sensitivity (Crowe & Matthews, 1964; Boyd, 1980; Matthews, 1981b). With γ_S stimulation the dynamic position sensitivity approximately equals the static position sensitivity (Matthews, 1981b). Stimulation of a γ_S is effective at preventing the silencing of the primary afferent during the release phase of the ramp stretch (Matthews, 1972, 1973). The dynamic responsiveness of the secondary afferent remains low during fusimotor stimulation (Boyd, 1980; Hulliger, 1984).

To small sinusoidal muscle stretches in the linear response range and at functionally relevant frequencies (less than 20 Hz), the sensitivity of the primary afferent is decreased by either γ_D or γ_S stimulation. The reduction of sensitivity by γ_S stimulation is much larger, falling to less than 20% of the value with no fusimotor stimulation, compared to a decrease to 70% with γ_D stimulation (Goodwin et al, 1975). The decrease in sensitivity to small

stretches when they are superimposed on a gradual stretch is prevented by γ_D activity. With γ_D stimulation, the responsiveness to sinusoidal stretches of large amplitude (up to 3 mm) and low frequency (less than 4 Hz) exceeds the passive value. At a frequency of 1 Hz, the amplitude at which γ_D stimulation increases the modulation is about 0.2 mm and it decreases with increases in frequency (Hulliger et al, 1977a). With γ_S stimulation, the decrease in modulation relative to that of the passive ending (when no γ-motoneuron stimulation is applied), is less for large stretches than small stretches (Hulliger et al, 1977a). The effect of γ_S stimulation on secondary afferents during small amplitude stretches over a range of frequencies is to decrease the sensitivity. The reduction is smaller than for primary afferents (Cussons et al, 1977). A decrease in the stretch modulation of the secondary afferent activity has also been found for large amplitude sinusoidal stretches at 1 Hz (Cussons et al, 1977). However, stimulation of some static γ-motoneurons has resulted in an increase in sensitivity to large amplitude stretches (Jami & Petit, 1981). Secondary endings are not often influenced by γ_D stimulation (Matthews, 1981b).

3.2.4 MECHANISMS OF FUSIMOTOR ACTIONS

How the functionally defined static and dynamic γ-motoneurons achieve their actions can be attributed to differences in the properties and innervation of the intrafusal fiber types. Such differences have been deduced

from histological and physiological experiments on the isolated muscle spindle.

The bag_1 fiber is unique because it exhibits mechanical creep. Following an extension, which is maintained, the sensory spirals creep back, shortening, over a 1 to 2 second period, and the firing rate of the primary afferent declines in parallel. This phenomenon is presumably due to a temporary increase in stiffness at the poles, induced or facillitated by the stretch. The stiffness then decays at the end of the movement, gradually decreasing the extension that is transferred to the sensory spiral, as the poles gradually lengthen the same amount. The increase in stiffness may result from an enhancement of cross-bridge formation by the stretch, either directly, or indirectly mediated by stretch-induced membrane depolarization (Boyd, 1980, 1983; Matthews, 1981a, 1981b; Hulliger, 1984). This characteristic of the bag_1 fiber can account for the difference, between primary and secondary afferents, in the dynamic sensitivity to large stimuli. Any terminals from a secondary afferent on the bag_1 fiber are located on a region with more myofilaments and so would not be affected in the same way (Banks et al, 1981).

Activity in a dynamic γ-motoneuron innervating the bag_1 fiber results in increased stiffness of the fiber during the stretch, so that the central sensorial region is extended by a greater amount than occurs in a passive fiber and the ensuing creep is larger. The increased stiffness of the active bag_1 fiber may be due to the attachment of cross-

bridges with a slow rate of turnover (Matthews, 1981a; Hulliger, 1984). This can explain why γ_D activity augments the dynamic response of the primary afferent (Boyd, 1980, 1983; Matthews, 1981a, 1981b).

A slow local contraction occurs in the polar region of the bag_1 fiber in response to stimulation of a γ_D axon. This extends the primary spiral ending by only about 5% and is responsible for the small increase in impulse rate under static conditions (Matthews, 1981a; Boyd, 1983).

Stimulation of a γ_S axon causes a strong fast local contraction of the bag_2 fiber which extends the primary spiral ending by about 20% and is responsible for the large bias in firing rate at any muscle length. When the muscle is simultaneously stretched, further extension of the spiral ending is limited, probably by the stiffness of the extended central region. Possibly for this reason, and because of the more rapid rate of cross-bridge turnover and the lack of creep, during γ_S stimulation the modulation in rate produced by sinusoidal stretch is relatively small and the dynamic position sensitivity of the primary afferent is similar to the static position sensitivity (Boyd, 1980, 1983).

Fast contracting chain fibers respond with propagated action potentials and twitch contractions to γ_S stimulation. The resulting twitch extensions of the primary ending account for the driving of the afferent when the activated γ_S axon innervates the chain fibers. The variation in γ_S action on the sensitivity of secondary endings could partly

be due to differences in the type and number of intrafusal fibers innervated by the γ-motoneuron (Jami & Petit, 1981).

3.2.5 FUNCTIONAL IMPLICATIONS OF MUSCLE SPINDLE CONTROL

The primary ending encodes some combination of length and velocity information. Because of the high sensitivity to movement of this receptor, the central nervous system (CNS) can quickly be made aware of a developing length change and so small perturbations can be rapidly corrected. This dynamic responsiveness is regulated by the fusimotor system; it is set to a high level by γ_D activity and to a low level by γ_S activity. During combined γ_S and γ_D activity, the primary afferent is moderately sensitive to large movements, but is relatively insensitive to small movements (Hulliger et al, 1977b). The primary afferent can be prevented from falling silent during muscle shortening by γ_S activity and also, but not as effectively, by γ_D activity under some conditions (Morgan et al, 1984).

The secondary ending, because of its low responsiveness to dynamic stimuli and consequently its ability to follow length decreases as well as increases, seems suited for providing the CNS with continual information pertaining to muscle length. Its relatively low sensitivity allows it to signal a wide range of muscle lengths without becoming saturated.

3.3 MUSCLE SPINDLE AFFERENT ACTIVITY AND FUSIMOTOR CONTROL DURING LOCOMOTION

The activity of single muscle spindle afferents during locomotion was first recorded in mesencephalic cats (Severin et al, 1967). Primary afferents from an ankle extensor fired at high rates not only during F, but more especially, during E2 and E3 (Severin et al, 1967; Severin, 1970). The occurrence of fusimotor activity, concurrent with skeletomotor activity, was required to explain the high firing rate during stance, when the active muscle lengthened briefly and then shortened (Severin, 1970).

In the decorticate cat, α-γ coactivation was demonstrated in tibialis anterior and gastrocnemius of the hindlimb (Perret and Buser, 1972), and in biceps bracchi, triceps bracchi, subscapularis, and teres major of the forelimb (Cabelguen et al, 1984). With the muscle held isometric, the primary afferent rate in each of the forelimb muscles was modulated in phase with the homonymous EMG (Cabelguen et al, 1984). For the experiments with gastrocnemius and tibialis anterior, in which the contralateral antagonist EMG was used for comparison, the afferent rate was strongest in phase with this activity, at about the time of the isotonic contraction of the homonymous muscle. The modulation in rate was greater for the ankle flexor (Perret and Buser, 1972).

In the acute spinal cat administered Dopa, simultaneous activation of alpha and gamma motoneurons, in both ankle

flexors and extensors, was inferred from studies of the primary afferent activity during stepping movements, and from the fusimotor activity during fictive locomotion (Sjostrom & Zangger, 1975, 1976). Thus the spinal central pattern generator for locomotion can control the activity in gamma, as well as in alpha motoneurons.

The investigation of muscle spindle activity during normal locomotion had to await the development of single unit recording techniques suitable for chronic use in the intact, freely moving cat (Prochazka et al, 1976; Loeb & Duysens, 1979). A primary afferent from an ankle extensor generally fired at a high rate during F followed by a much reduced rate in E1. A burst of impulses at the time of foot contact decayed to a moderate but decreasing firing rate in E3. The afferent firing rate during the step cycle was thus predominantly related to changes in muscle length (Prochazka et al, 1976, 1977). However, during E1, E2, and E3 the afferent was more active than during manually replicated length changes with the cat functionally de-efferented by deep barbiturate anesthesia. A fairly moderate level of γ-motoneuron activity, possibly both static and dynamic, was concluded to occur concomitantly with the α-motoneuron activity in ankle extensors. The higher firing rate observed during the support phase for the mesencephalic preparation, relative to the intact cat, could have resulted from the effects of hip fixation in the mesencephalic cat (Prochazka et al, 1977). The firing rate of a secondary afferent from an ankle extensor closely reflected the muscle length

throughout the step cycle (Loeb & Duysens, 1979).

The primary afferents studied from ankle flexors (Loeb & Duysens, 1979) fired more quickly during rapid shortening of the active muscle in the F phase than during passive lengthening in the E1 and E3 phases. Strong modulated activity in γ_S motoneurons accompanying the activity in α-motoneurons could explain this (Loeb & Duysens, 1979).

Afferents from muscles controlling the other joints have shown diverse behaviours (Prochazka et al, 1976; Loeb & Duysens, 1979; Loeb, 1984). The type, timing, and degree of fusimotor activity in a muscle may depend on the function of that muscle in locomotion (Loeb & Duysens, 1979; Loeb, 1984). For example, α-γ coactivation involving static γ-motoneurons in flexors and dynamic γ-motoneurons in extensors has been suggested (Loeb & Hoffer, 1981).

Three different approaches have been used to try to more clearly determine the relative contributions of the static and dynamic systems to the fusimotor activity during walking. Afferent responses to applied stretches were examined during locomotor activity in the decorticate cat. In the first of these experiments (Perret and Berthoz, 1973), because the muscle stretcher was not controlled by a displacement servo, observations were only made in the absence of contraction of the muscle from which the afferent activity was recorded. In response to 4 mm sinusoidal stretches, the primary afferents in gastrocnemius showed a cyclic increase in their dynamic response, while afferents

in tibialis anterior showed a cyclic decrease in the stretch induced modulation at a high mean rate. These results were interpreted as being due to a phasic γ_D action in the ankle extensor and a dominant phasic γ_S action in the ankle flexor. More recently, (Cabelguen, 1981), responses to sinusoidal and ramp stretches were examined in experiments in which muscle contraction was allowed to occur. One hindlimb was fixed and the muscle of interest held isometric, except for controlled stretches. The cat walked on a treadmill with the other three legs. The previous conclusions were confirmed and extended: a strong phasic γ_D action accompanied by a weak γ_S action occurred in gastrocnemius spindles, and a weak dynamic but strong static action in phase with the EMG was claimed for spindles in tibialis anterior. When the cat was not walking, one gastrocnemius afferent showed the possibility of a γ_D, with perhaps a weak γ_S, influence. For the proximal bifunctional muscles, the static and dynamic fusimotor actions were balanced.

However, Cabelguen's experiments (1981) suffered from several limitations. The interpretation from the primary afferent recordings of the type and strength of fusimotor activity can not be assumed to be unequivocal. Experiments on anesthetized cats have shown that static γ-motoneurons can have a variable effect on the dynamic response to large ramp stretches (Brown et al, 1965). In combination, the action of one type of γ-motoneuron may at times be occluded by the action of the other type (Hulliger et al, 1977b).

Maintained spindle afferent discharge during muscle shortening is not necessarily indicative of a static γ-motoneuron action since γ_D stimulation has also achieved this (Morgan et al, 1984). Secondly, the long period of the applied length changes limited the number of different times within the step cycle period that the spindle response could be determined. The variability of the step cycle period prevented reliable correlation of spindle responses with EMG activity. Furthermore, the effect on the spindle response of internal shortening of the extrafusal fibers during the isometric muscle contractions was ignored.

Recently, a simulation technique has been used to deduce the fusimotor activity in the soleus muscle during cat locomotion (Appenteng et al, 1983; Hulliger et al, 1984; Prochazka & Hulliger, 1983). In anesthetized cats, the muscle length and tension changes of locomotion were cyclically reproduced with different patterns of γ-motoneuron stimulation. The spindle behaviour was compared to that during normal walking. Satisfactory matches were obtained sometimes with tonic γ_S stimulation, sometimes with tonic γ_D stimulation, and occasionally with combined tonic and EMG-modulated stimulation patterns. However, in no case was a satisfactory match of the experimental records made without any γ-motoneuron activity or with modulated stimulation alone used in the simulation. From these early results, the occurrence of tonic γ-motoneuron activity, and hence some independence of alpha and gamma activation during

normal walking, was concluded. Very recently, in a case in which tonic γ_S stimulation provided a good match, the addition of modulated γ_D stimulation was found to maintain the good match unless the modulation was very large (Hulliger, personal communication).

Static and dynamic fusimotor activity has had to be deduced from single afferent recordings due to the difficulty of recording from single, functionally identified, γ-motoneurons during locomotion. Because such an indirect method relies on various assumptions concerning the spindle response, the inferred γ_D and γ_S activity has remained inconclusive and controversial.

However, the challenge of recording directly from single identified γ-motoneurons during locomotion has recently been met (Stein et al, 1983; Murphy et al, 1984). Premammillary cats walked with three legs on a treadmill. The fourth limb was fixed in place and dissected single γ-motoneurons were recorded from cut triceps surae nerves. The firing rate of one type of gamma axon was low at rest, 10 $\pm$ 6 imp./s (mean $\pm$ S.D.). During walking, the firing rate was markedly increased, to 56 $\pm$ 10 imp./s, but showed relatively little modulation (13 $\pm$ 5 imp./s) within the step cycle. The other type of gamma axon had a higher firing rate at rest, 44 $\pm$ 16 imp./s. The mean rate during walking, 36 $\pm$ 9 imp./s, was not significantly different from the resting discharge. However, the rate was highly modulated (27 $\pm$ 8 imp./s) with each step (Murphy et al, 1984). The fusimotor axons were termed tonically-modulated, γ_t, and

phasically-modulated, γ_p, respectively. The peak γ_t firing rate preceded the peak EMG, whereas the peak γ_p activity occurred about one-third of the step cycle later, near the time of peak tension.

The correspondence of tonically-modulated and phasically-modulated axons with static and dynamic γ-motoneurons was determined in other cats using nerve branches and ventral roots maintained in continuity (Stein et al, 1983; Murphy et al, 1984). Identification of static and dynamic γ-motoneurons was achieved by recording the primary afferent activity in fine branches of the LG or MG nerves. The stretch responses, with and without ventral root stimulation of a functionally single γ-motoneuron affecting the spindle, were compared. The resting firing rate of each identified single γ-motoneuron was then recorded from the nerve branch following section of the other ventral root filaments and of the muscle nerve distal to the recording site. The six static γ-motoneurons studied all had a low firing rate and the two dynamic γ-motoneurons both had a high firing rate, indicative of γ_t and γ_p axons, respectively. From this and other more indirect evidence we concluded that the tonically modulated pattern of activity occurred in static γ-motoneurons, and the phasically modulated pattern of activity occurred in dynamic γ-motoneurons. Since γ-motoneurons influence the bias and sensitivity of the muscle spindle, the functional implications of these findings must be ascertained by direct

recordings from muscle spindles during locomotion in the premammillary cat.

This challenge provided the impetus for this thesis. Because spindle activity is determined in a complex manner by length changes and fusimotor drive, the muscle was held isometric to separate the fusimotor effects from the effects of the locomotor length changes. Controlled stretches were applied to the muscle to determine the sensitivity of the spindle to stretch at different times in the step cycle. Since secondary afferents are only affected by static γ-motoneurons and primary afferents are affected by both static and dynamic γ-motoneurons, their stretch responses were compared to provide additional evidence for the identification of the fusimotor activity patterns described for the premammillary cat. To determine the effect of static and dynamic γ-motoneurons on the mean rate and sensitivity of primary afferents, single identified γ-motoneurons were stimulated at steady rates in anesthetized cats while applying muscle stretches and recording from single primary afferents. Pairs, consisting of a static and a dynamic γ-motoneuron, were also stimulated with the rate patterns observed during walking. These experiments in anesthetized cats were done to provide a basis for interpretation of the changes that occurred during walking.

CHAPTER FOUR

STRETCH RESPONSES OF MUSCLE SPINDLE AFFERENTS DURING LOCOMOTION

4.1 METHODS

4.1.1 EXPERIMENTAL PROCEDURE

Forty-one adult cats were used in acute experiments for the investigation of muscle spindle responses during locomotion. The cats, of either sex and weighing 2.0 to 5.0 kg, were anesthetized with halothane delivered in gaseous form mixed with 95% O_2 and 5% CO_2. A tracheotomy was done, a common carotid artery was cannulated for monitoring blood pressure, and an external jugular vein was cannulated for administering dextran to compensate for blood loss. The other carotid artery was ligated to reduce blood flow to the brain and hence minimize bleeding from the effects of later surgery.

A laminectomy was done to expose the L_7 and S_1 dorsal roots which contain muscle spindle afferents from soleus. To reduce the activity of afferents from other muscles which travel in the L_7 and S_1 dorsal roots, the muscles of the left hindlimb and tail, excluding soleus, were denervated. The soleus muscle of the left hindlimb was freed from the surrounding muscles and its tendon detached along with a small piece of the calcaneus. An EMG electrode was sutured to the muscle. In early experiments, a silastic cuff electrode was placed around the soleus nerve. Since this

cuff sometimes blocked the nerve, hook electrodes were later used instead.

The cat was mounted in a stereotaxic frame over a treadmill. A vertebral clamp and pins at the iliac crests held the cat in position and partially supported its weight. Knee pins held the left hip slightly extended and the knee flexed. A foot clamp allowed the soleus muscle to be immersed in a pool of paraffin oil. An oil pool was also formed around the spinal cord. Parts of the L_7 and S_1 dorsal roots were cut and reflected for later recording. The temperature of the oil pools was maintained near 37.5°C with radiant heat. Local anesthetic, Novocaine, was applied to the cut skin surfaces and injected around the hip bars to minimize pain sensation.

The cat was reduced to a premammillary preparation. To do this, the cortex was removed and the brainstem was sectioned from just rostral to the superior colliculus to just rostral to the mammillary bodies. Gas anesthesia was discontinued. The perception of pain is thought to be absent in the premammillary cat as it is in other decerebrate preparations. The advantage of the premammillary preparation is its ability to spontaneously initiate periods of locomotion. We favour the premammillary preparation over the mesencephalic preparation, which can produce locomotor activity in response to appropriate electrical stimulation, because we have found walking in the former preparation to be more reliable and regular. Also, there is no stimulus artefact to interfere with the recorded

signal.

Functionally single muscle afferents were dissected from the cut dorsal roots and recorded on hook electrodes. The responses to twitch contractions elicited by stimulation of the soleus nerve were used to distinguish muscle spindles from tendon organs. Conduction velocity, and the response to large ramp stretches, were used to identify a spindle afferent as a primary (CV: 75-117 m/s, n=21) or a secondary (CV: 23-69 m/s, n=8). Conduction velocity was calculated from measurements of the distance between the soleus nerve and dorsal root electrodes, and the conduction latency when the soleus nerve was stimulated. The values for conduction velocity were corrected to 37.5°C using a Q_{10} of 1.32 (Franz & Iggo, 1968).

The soleus tendon was connected to a strain gauge, for tension measurement, in series with a torque motor. A potentiometer attached to the torque motor provided a measurement of muscle length. Negative length and velocity information was fed back to the torque motor. This allowed the torque motor to be used to apply controlled sinusoidal or ramp stretches to the muscle, or to hold the muscle at constant length. The mean muscle length was adjusted to the length at which the twitch tension was maximum. The sinusoidal stretches were of 4 and 10 Hz, and between 0.15 and 0.72 mm in amplitude (1/2 peak to peak). The ramp stretches and releases were applied at a constant velocity between 15 and 20 mm/s over a distance between 1.5 and 1.7

mm. The period of one cycle was 600 ms.

The cat walked with three legs on the moving treadmill belt. Stepping movements usually started when the treadmill was turned on and stopped when it was turned off. Appropriately timed bursts of activity occurred in the soleus muscle of the fixed hindlimb. Sinusoidal and ramp stretches were applied to the soleus muscle while the cat was walking and also while it was standing quietly. During the period of locomotion, the stretches occurred at all phases of the step cycle.

The tension, length, and EMG signals, and the neural activity from the dorsal root filament were processed, monitored on oscilloscopes and a pen recorder, and along with a marker for the stretch cycle, recorded with an FM tape recorder. The initial processing entailed suitable amplification of the signals for the 5 volt range of the tape recorder. Before recording on tape, the EMG signal was high pass filtered to remove the movement artefact. The neural activity was also high pass filtered to improve the signal to noise ratio by reducing the powerline interference.

4.1.2 ANALYSIS

Offline, the EMG signal was rectified and smoothed with low pass, RC (50 Hz) and Paynter (30 Hz) filters (Gottlieb & Agarwal, 1970). This signal was used to produce step cycle markers from a Schmitt trigger. So that only one pulse occurred per cycle, the duration of the pulse that was

triggered when the EMG exceeded a preset level, was adjusted to be longer than the burst of EMG activity. A brief marker was generated at the start of each trigger pulse.

The neural signal was bandpass filtered, if necessary, so that the identified afferent could be reliably distinguished from the background noise and any other afferent activity. An acceptance pulse was produced on each occurrence of the signal through an adjustable amplitude-time window. In this manner, the analog spike train was represented digitally.

Fig. 1 shows a typical example of the processed signals and markers fom a period of locomotion during which 4 Hz sinusoidal stretches were applied. Although the EMG signal was shifted in time with respect to the other signals as a result of the filtering, this lag was short (20-25 ms) compared to the duration of the step cycle (greater than 500 ms). It is readily apparent that the frequency of the sinusoidal stretch was not synchronized with the frequency of the stepping, so that during a walking sequence the stretch occurred at all times in the step cycle. The sinusoidal length change slightly modulated the force produced by the muscle. The system had some compliance so that the sinusoidally varying muscle length was influenced cyclically by the muscle contractions. However, the variations in amplitude of the sinusoidal length change were sufficiently small (less than 0.02 mm) so as to be of little consequence.

Averaging of analog signals, and the generation of

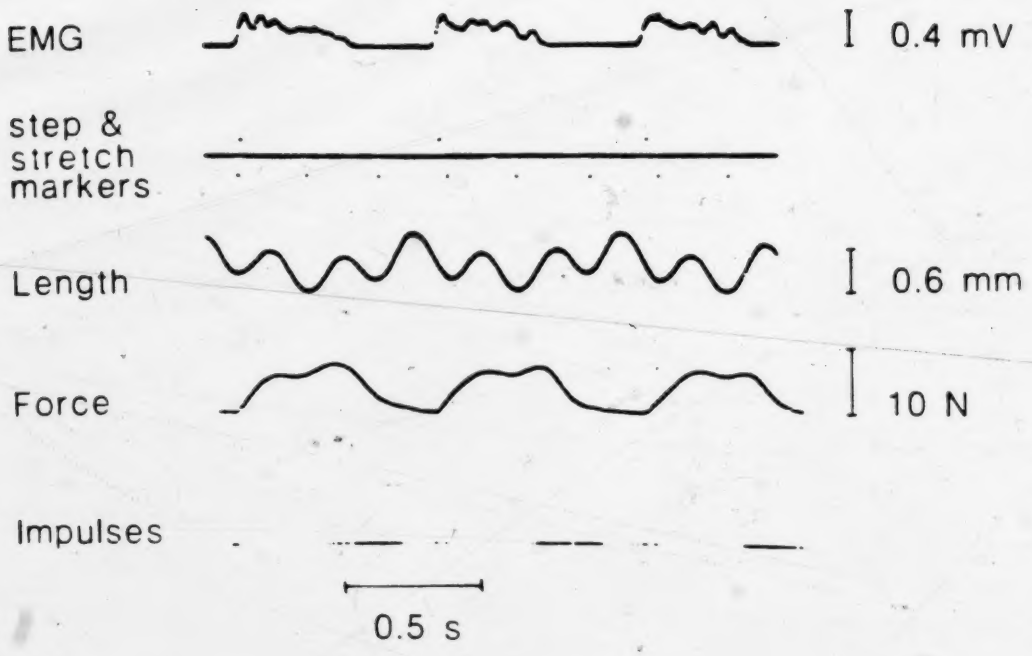


Figure 1. Recorded signals from a period of walking. The rectified and filtered soleus EMG was used to generate markers of the step cycle. The muscle was stretched sinusoidally at 4 Hz and stretch cycle markers were recorded. The step and stretch cycle markers were used to trigger the computer sweeps for the step cycle and stretch cycle averages, respectively. The actively generated muscle force affected the resulting length change and the sinusoidal stretches affected the muscle force. The impulses of the primary afferent are shown here as dots.

cycle histograms from the afferent pulses, was achieved with a PDP 11/34 computer. To generate the cycle histogram, the afferent impulses in each of the bins following the trigger pulse were counted. The number of pulses was divided by the number of sweeps and the bin width, so that the average afferent activity could be displayed as firing rate, in units of impulses/s. Although this is referred to as a cycle histogram, it is more correctly termed a probability density function. The value in each bin was averaged with the values in the two bins on either side. This five point running average reduced the scatter, resulting in a smoother histogram.

To determine the response of the afferent to the applied length changes, the stretch markers were used to trigger the sweeps of the computer. The spans of the stretch cycle histograms and the length and tension averages, which were computed simultaneously, were set approximately equal to the period of the applied stretch. An example is illustrated in Fig. 2. The afferent response to sinusoidal stretch was fitted with a curve by the method of least mean squares (Sokolnikoff & Redheffer, 1966) using the equation $y = a \sin(2\pi t/T) + b \cos(2\pi t/T) + c$. In this equation, y is the afferent response at any time, t, to a stretch of period, T. The amplitude, $\sqrt{(a^2 + b^2)}$, where a and b are the sine and cosine coefficients, and the mean, c, of the fitted curve, were used as measures of the modulation in rate and the mean impulse rate. The modulation in rate

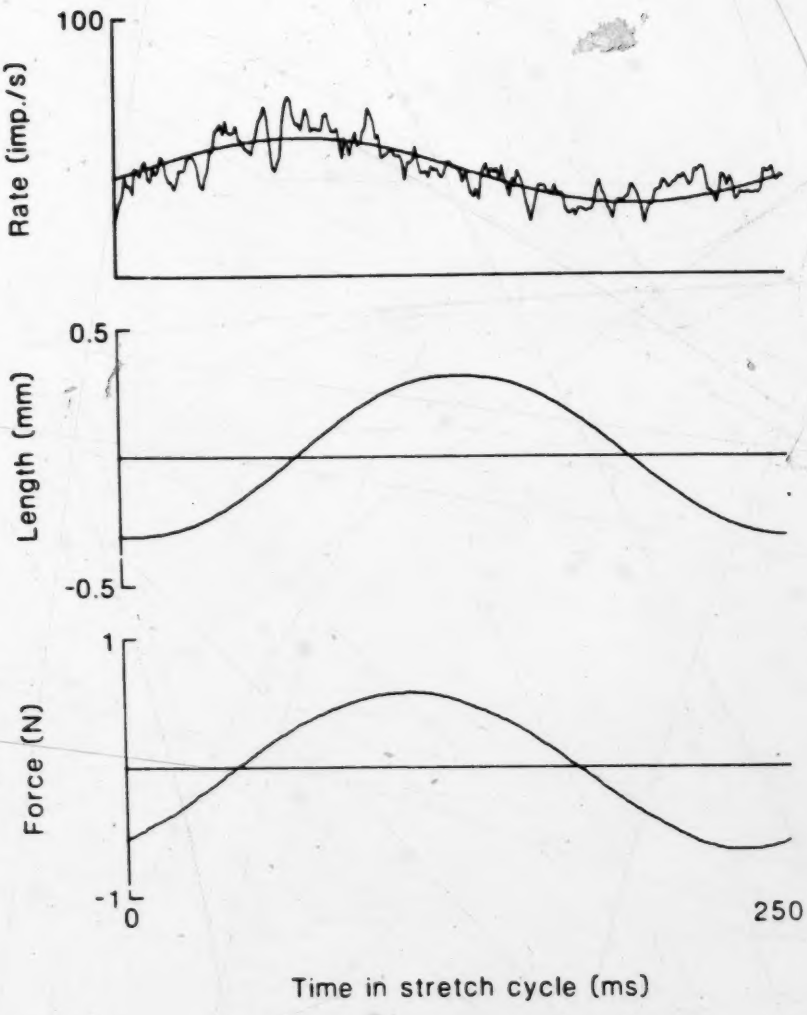


Figure 2. Stretch cycle averages. The response of a primary afferent to 4 Hz, 0.31 mm sinusoidal stretch during walking was averaged, and the smoothed histogram was fitted with a curve. Length and force were averaged simultaneously, with each computer sweep triggered by a stretch cycle marker.

was expressed in terms of sensitivity by dividing the amplitude of the fitted sine by the average amplitude of the length change. Because of the nonlinear behaviour of the afferent, and the different amplitude of the applied length change in different experiments, per cent changes in sensitivity were compared, rather than differences in sensitivity directly.

To assess the muscle spindle activity during the step cycle with respect to the EMG and tension changes, each sweep of the computer was triggered by a step cycle marker. The EMG and tension signals were averaged concurrently with the computation of the step cycle histogram. This is illustrated in Fig. 3 for the same primary afferent that is shown in Fig. 2 and during the same period of walking. The bin width was usually selected so that the 256 bins of the sweep included slightly less than one step cycle. As much of the step cycle as possible was shown without greatly reducing, due to the processing time of the computer, the number of steps averaged.

To examine how the sensitivity to stretch varied at different times within the step cycle, a more complicated method of analysis was required. The step cycle was divided into 14 parts and the afferent response to sinusoidal length changes was determined for each part. Histograms, originating at the time of the stretch marker and extending over one step cycle, were computed from the afferent responses to all the stretches which began in the same one-fourteenth of the step cycle. In Fig. 4A, the first

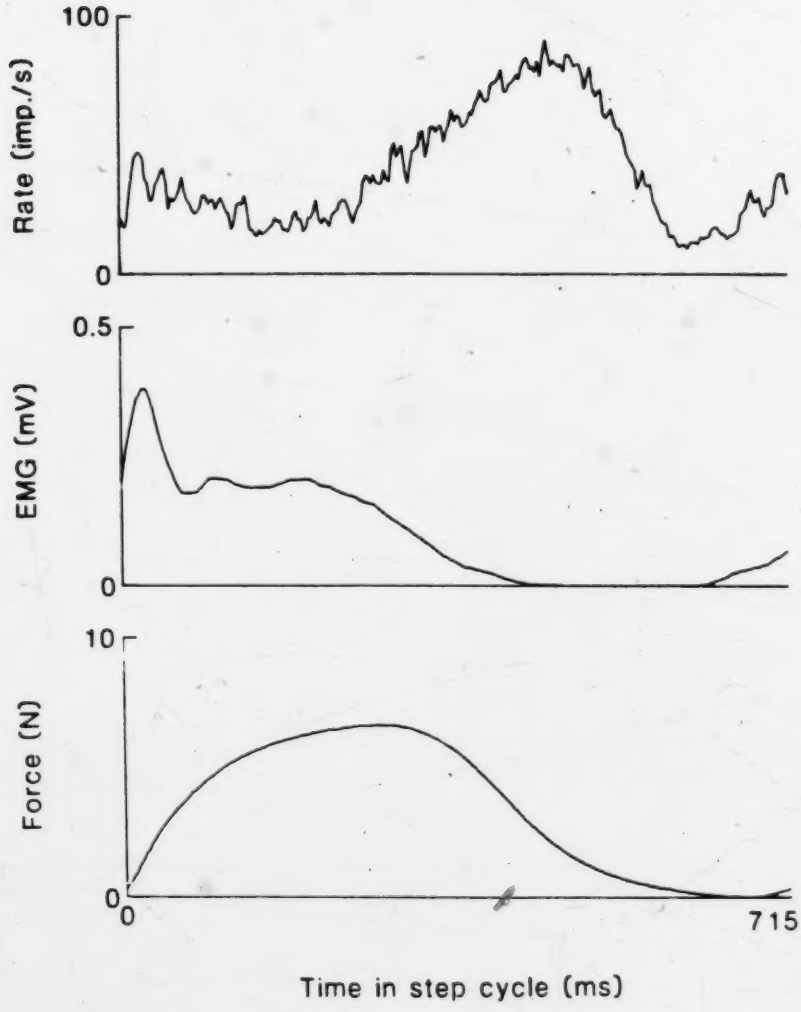
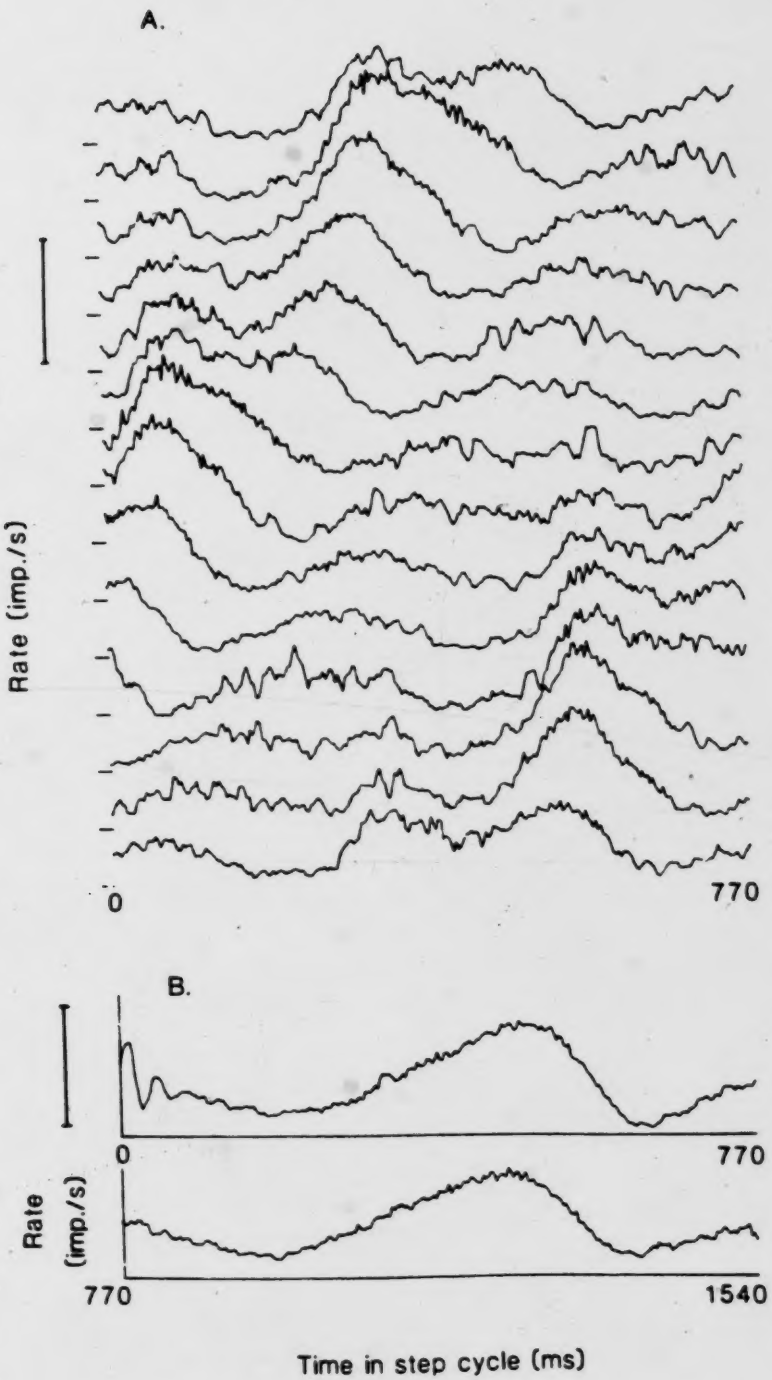


Figure 3. Step cycle averages. The activity of the same primary afferent as in Fig. 2 was averaged over the step cycle and then smoothed. Step cycle markers triggered the sweeps for the histogram and also for the averages of the EMG and force. The traces span 715 ms, which is slightly less than the mean period of the step.

Figure 4. A. Separate histograms of the impulse rate of a primary afferent for 14 phases of the step cycle. Each starts at the time of the stretch cycle marker. The stretches began 1/14 of the step cycle (54 ms) later for each histogram. Each histogram extends over the period of one step and so includes the response to about 3 cycles of stretch. B. Step cycle histogram which extends over 2 step cycles. The appropriate portion of it, 1 step cycle in duration, can be subtracted from each of the 14 histograms in A. Calibration bar = 100 imp./s.


A.
Rate (imp./s)
0
770
B.
0
770
Rate
(imp./s)
770
1540
Time in step cycle (ms)

histogram shows the afferent activity when stretches began 0-54 ms after the step cycle marker. The stretches started after the step markers by increments of 54 ms for each successive histogram. For example, the seventh and fourteenth histograms are the average afferent responses for stretches beginning 324 to 378 ms, and 702 to 756 ms, respectively, after the start of the step. The histograms in Fig. 4A were dependent on both the locomotion induced activity and the stretch induced activity. The mean firing rate, which reflected both of these influences, could be obtained from the portion of each histogram corresponding to one stretch cycle. If plotted against time in the step cycle, the resulting representation should resemble the overall step cycle histogram. However, to obtain the modulation in afferent rate due solely to the stretch, the average response over the step cycle was subtracted from each histogram. Because each of the fourteen histograms started at a different time in the step cycle, the step cycle average (Fig. 4B) had to be shifted appropriately before it was subtracted. This was possible because the overall average in this procedure was computed over two step cycles although the bin width was still equal to that for the fourteen histograms. Following subtraction, the spans of the fourteen histograms were adjusted so that just one stretch cycle was included. Each of the histograms was then fitted with a sine curve (Fig. 5) and the modulation in rate determined. In this manner, the responses at different

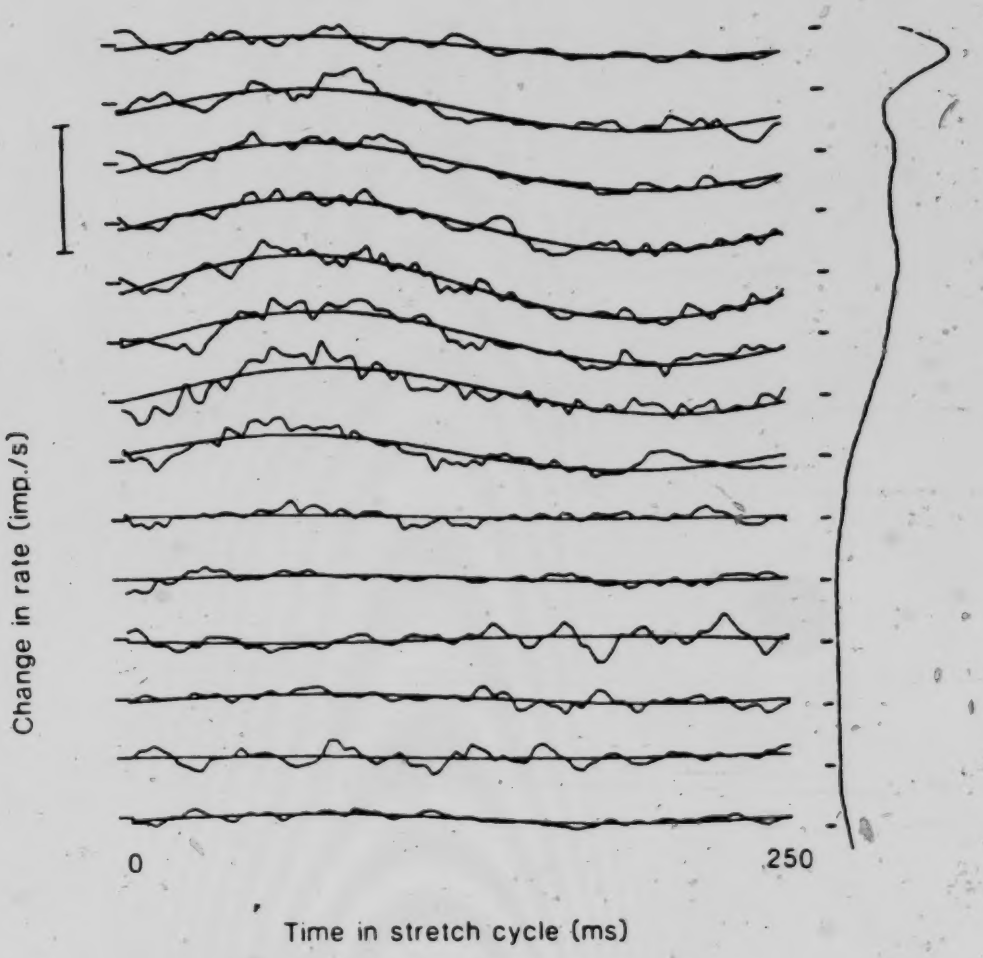


Figure 5. Curve fitted, stretch cycle histograms for 14 phases of the step cycle. The average activity during the step (Fig. 4B) was subtracted from each of the traces in Fig. 4A to leave the afferent response to the stretch alone. The response to only 1 cycle of stretch is illustrated. Calibration bar = 100 imp./s. The EMG is shown on the right and the time of the beginning of each of the 14 phases is indicated. The time in the step cycle increases from top to bottom.

times in the step cycle could be compared. A similar analysis, without the subtraction procedure, coud be used on the length signal to determine the amplitude of the stretch in each of the fourteen parts. Because the variation in amplitude was found to be small, this was not usually done, and the results were not distorted when instead the average length change was used to calculate sensitivity.

Ramp stretches were not used to study changes within the step cycle because of the long duration, 600 ms, of one stretch cycle.

The validity of the foregoing procedure was checked using known inputs to an electronic neural analog. The neural analog was adjusted to generate a steady train of impulses at 40 imp./s. This rate was modulated by two sinusoidally varying inputs. One, a 4 Hz sinusoid, was meant to imitate the effect of stretches, and modulated the mean rate by $\pm$ 10 imp./s. The other, a 1.5 Hz sinusoid, was meant to imitate the effect of stepping on the mean rate, and modulated it by $\pm$ 30 imp./s. The steady rate of the neural analog was modulated by the sum of the two sinusoids. The procedure explained above, for determining the stretch sensitivity at different times in the step cycle, was implemented. The modulation in rate was found to be the same, 10 imp./s, in all the fourteen histograms, so that the sensitivity was constant, as expected. Thus the subtraction of the overall step cycle average, appropriately shifted, does indeed result in the rate modulation due to the stretch alone.

So that the sensitivity would vary, the two sinusoidal signals were multiplied before modulating the rate of the neural analog. In this case, subtraction of the step cycle histogam did not result in a constant modulation of impulse rate.

4.2 RESULTS

Twenty-one primary and eight secondary soleus muscle spindle afferents were studied from premammillary cats which walked with three legs on a treadmill. Controlled stretches were applied to the soleus muscle of the fourth limb. Although the length changes of stepping were prevented in that hindlimb, the soleus muscle nonetheless contracted with the walking rhythm. The EMG, force, length, and neural signals, and markers of the step and stretch cycles, are shown in Fig. 1.

4.2.1 AFFERENT RESPONSES WITHIN THE STRETCH CYCLE

Using the stretch cycle markers to average the signals, the afferent response to the applied stretch was determined. Fig. 2 shows the stretch cycle histogram for a primary afferent and the corresponding length and force averages. The response to 4 Hz sinusoidal stretch when the cat was standing quietly can be compared to that when walking, for a primary afferent in Fig. 6 and for a secondary afferent in Fig. 7. The mean rate increased from 22 imp./s at rest to 41 imp./s during walking for the primary afferent, and from 20

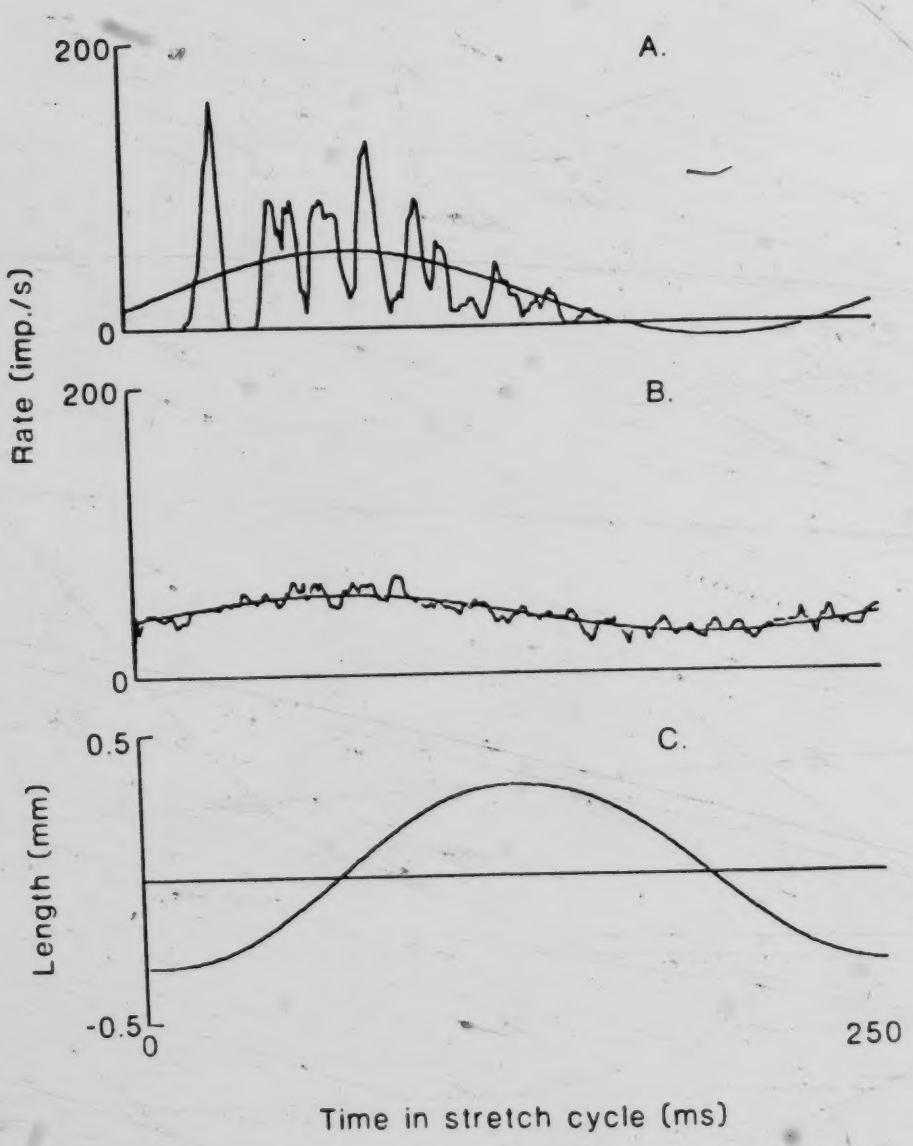


Figure 6. Responses of a primary afferent to 4 Hz, 0.3 mm sinusoidal stretch. The stretches were applied while the cat was at rest (A) and during walking (B). The sinusoidal length change is shown in C.

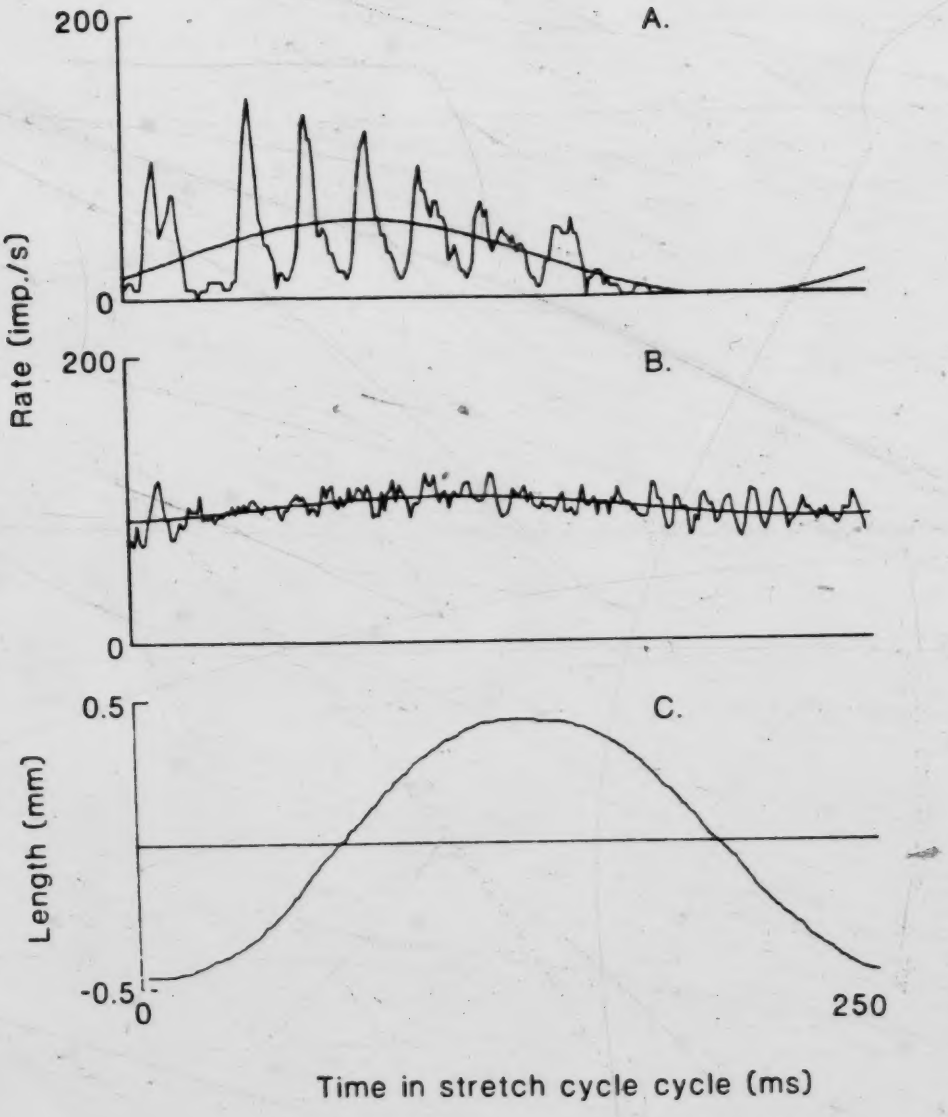


Figure 7. Responses of a secondary afferent to 4 Hz, 0.4 mm sinusoidal stretch. The stretches were applied while the cat was at rest (A) and during walking (B). The sinusoidal length change is shown in C.

imp./s to 91 imp./s for the secondary afferent. On average, the mean rate increased by 23 ± 17 imp./s for the primary afferents, and by 38 ± 25 imp./s for the secondary afferents. These increases were statistically significant at $p < 0.001$ for the primary afferents and $p < 0.01$ for the secondary afferents, using Student's t test. The modulation in rate of the primary afferent decreased from 32 imp./s in response to 0.33 mm stretches at rest to 14 imp./s for 0.31 mm stretches during walking. The modulation in rate of the secondary afferent to slightly larger stretches decreased from 23 imp./s at rest to 8 imp./s during walking. The sensitivity of the primary afferent thus decreased by 54%: from 97 imp. s^{-1} mm^{-1} to 44 imp. s^{-1} mm^{-1}. The sensitivity of the secondary afferent decreased by 61%: from 51 imp. s^{-1} mm^{-1} to 20 imp. s^{-1} mm^{-1}. For all the primary afferents studied, the sensitivity was reduced during walking by 49 ± 26% (statistically significant at $p < 0.001$). For the secondary afferents, the reduction was by 33 ± 36% (statistically significant at $p < 0.05$).

The responses to ramp stretches were similarly influenced during walking, as shown in Fig. 8 and Fig. 9. For both the primary and secondary afferent, the mean rate was elevated and the dynamic responsiveness was reduced during the period of walking, compared to the rest period.

Muscle spindle afferents are affected by length changes and fusimotor activity. Thus, the differences observed between the rest and walking conditions can be attributed solely to the differences in fusimotor drive only if the

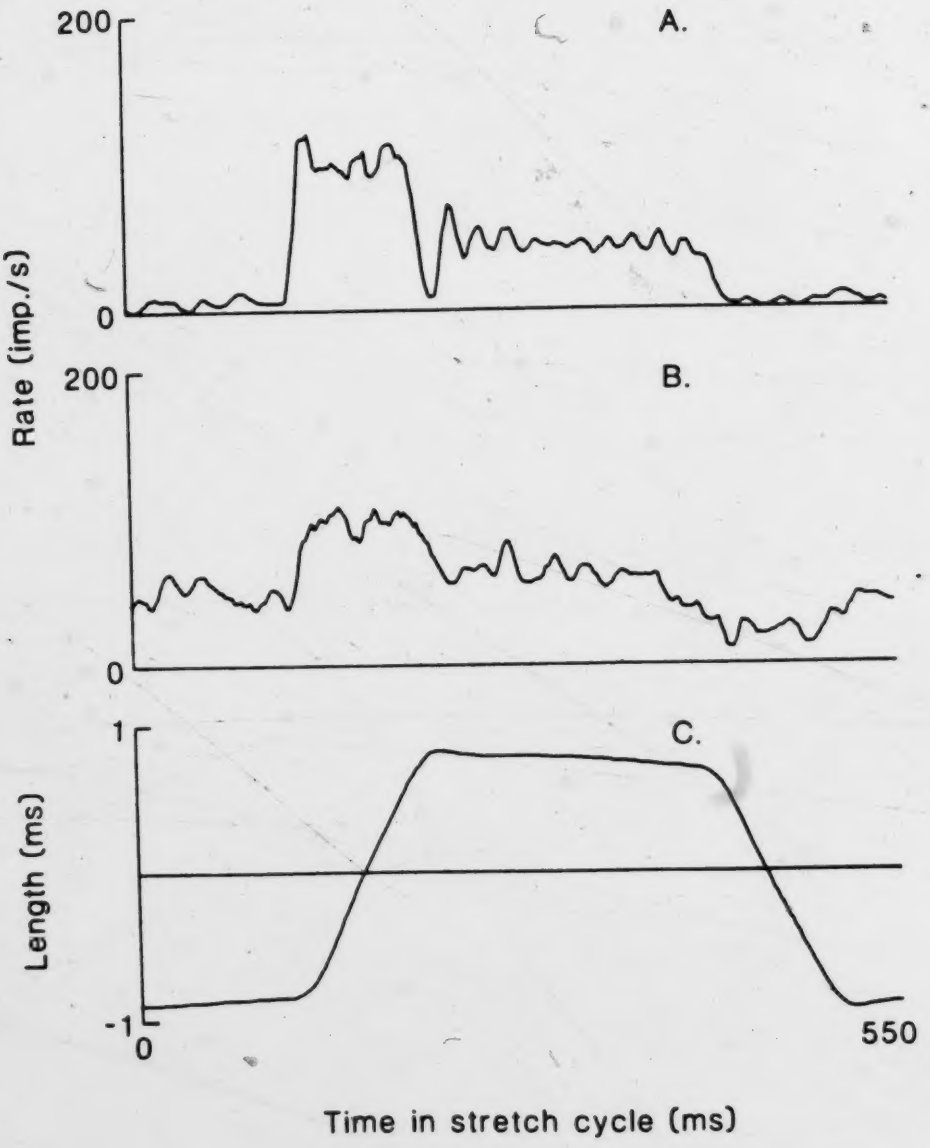


Figure 8. Average responses of a primary afferent to ramp stretches. The length changes (C) were applied while the cat was at rest (A) and during walking (B). The cycle period was 600 ms, the stretch amplitude was 1.6 mm and the velocity of the stretch was 16 mm/s.

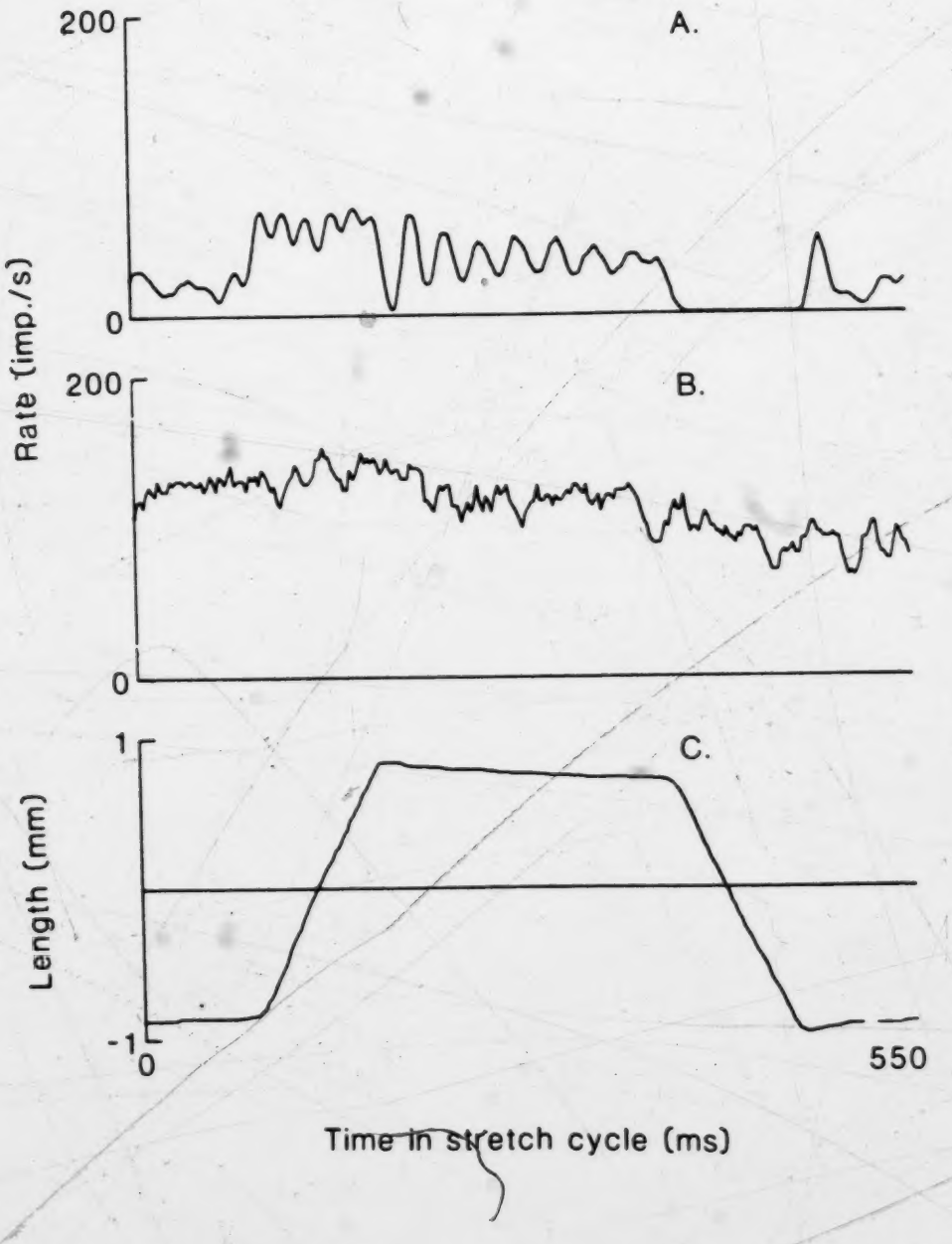


Figure 9. Average responses of a secondary afferent to ramp stretches at rest (A) and during walking (B). As shown in C, the stretch amplitude was 1.6 mm, the velocity of the stretch was 16 mm/s, and the cycle period was 600 ms.

length changes affecting the spindle were the same. Because the length was controlled by negative feedback, the external length change was relatively constant for the two experimental conditions, despite the quite different activation states of the muscle.

However, during walking, the muscle fibers shortened internally as the muscle contracted and regained their original length as the muscle relaxed. The muscle spindle would be affected by these internal changes. To estimate the amount of internal shortening that occurred during walking, the average muscle stiffness was calculated by dividing the amplitude of the stretch cycle-averaged force change by the amplitude of the stretch cycle-averaged length change. For example, in Fig. 2, this is the amplitude of the third trace, 0.60 N, divided by the amplitude of the second trace, 0.31 mm. The mean force over the step cycle, for example, the mean of the second trace in Fig. 3, 3.37 N, was divided by the average muscle stiffness, 1.9 N/mm, to give the average length by which the muscle was internally shortened during the step cycle. The internal shortening calculated for the walking period of this example was 1.8 mm. The internal shortening for all the walking sequences for which it was calculated was 2.9 ± 1.8 mm. The effect of this net reduction in muscle length would be to decrease the firing rate. Since the mean rate during walking was greater than at rest, a relatively strong fusimotor drive must have been present when the cat was walking.

4.2.2 AFFERENT RESPONSES WITHIN THE STEP CYCLE

Using the step markers, the EMG, tension, and afferent activity were averaged over one step cycle. Although small stretches were applied throughout the walking sequence, they occurred randomly with respect to the step cycle markers, and did not affect the step cycle histogram. In Fig. 10, the step cycle histograms from sequences of locomotion, in which stretches were (B) and were not (A) applied, are similar. Apparently, the effect of the rising phase of the stretch on increasing the firing rate was counteracted by the effect of the falling phase on decreasing it, so that the firing rate was not offset for the period of locomotion during which the stretches were applied. In this example, the walking was more forceful during the period in which the muscle was not sinusoidally stretched, and probably for this reason the modulation in rate during the step cycle was greater in B than in A.

The activity of both primary and secondary afferents was relatively low during the buildup of muscle force and increased as the force declined. On average, the peak firing rate occurred after the peak in force by 236 $\pm$ 160 ms (24 $\pm$ 12% of the step cycle) for the primary afferents studied, and by 339 $\pm$ 120 ms (37 $\pm$ 11% of the step cycle) for the secondary afferents. The peak in impulse rate at that time can largely be attributed to the internal length changes produced by the isometric muscle contractions. Examples in which the impulse rate can be compared to the muscle force and the rate of change of force, during the

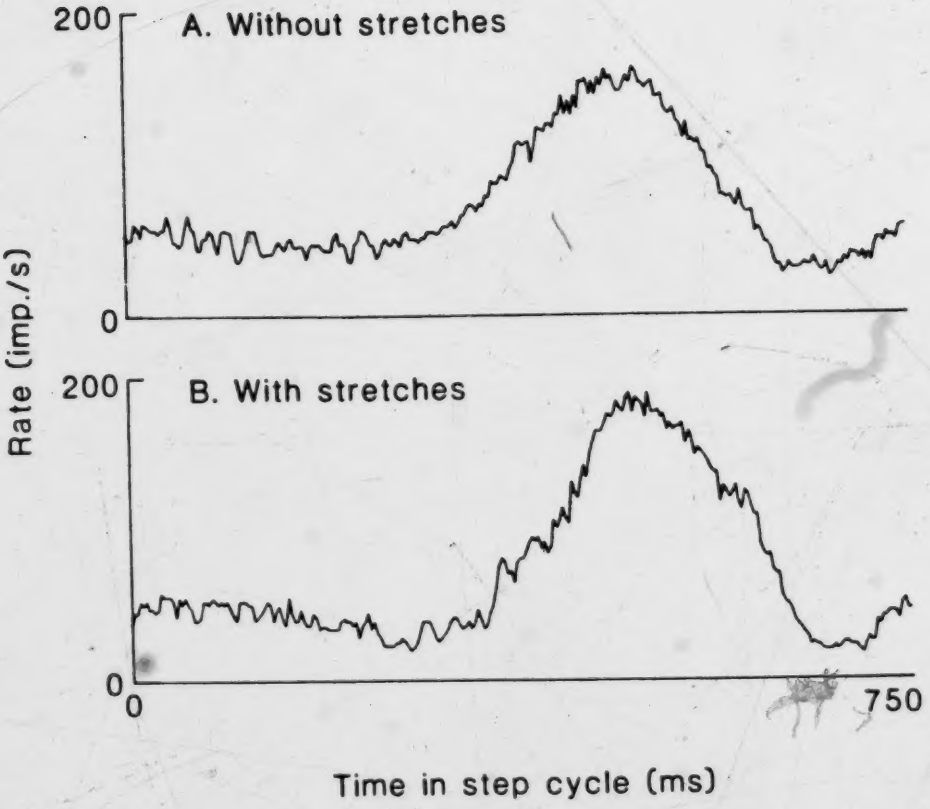


Figure 10. Average activity of a primary afferent during the step cycle when the muscle was held isometric (A), and when 4 Hz sinusoidal stretches were applied (B).

step cycle, are shown in Fig. 11A for a primary afferent and in Fig. 11B for a secondary afferent. As the muscle force increased, the extrafusal fibers shortened internally and unloaded the muscle spindle. The muscle spindle was stretched back to its original length when the force declined. Note the close inverse relation between the rate of change of force, which is associated with the velocity of shortening, and the impulse rate of the primary afferent. The primary afferent, which has greater dynamic sensitivity, shows a larger variation in rate than the secondary afferent.

The primary afferent in Fig. 3 shows a small peak in firing rate at the time of the onset of the EMG, but the primary afferent in Fig. 10 does not. The early peak in rate, present in 50% of the primary afferents studied, preceded the peak tension by 272 $\pm$ 263 ms (28% of the step cycle). Because the active force was low then, the early augmentation in rate can be explained by fusimotor activity at that time. Variability in the strength of the fusimotor influence could account for the observed differences among afferents.

4.2.3 SENSITIVITY CHANGES WITHIN THE STEP CYCLE

During the step cycle, the sensitivity to 4 Hz sinusoidal stretch was highly modulated for the primary afferents, compared to the secondary afferents. Examples of the changes during the step cycle in the sensitivity and mean rate are shown for a primary afferent in Fig. 12, and

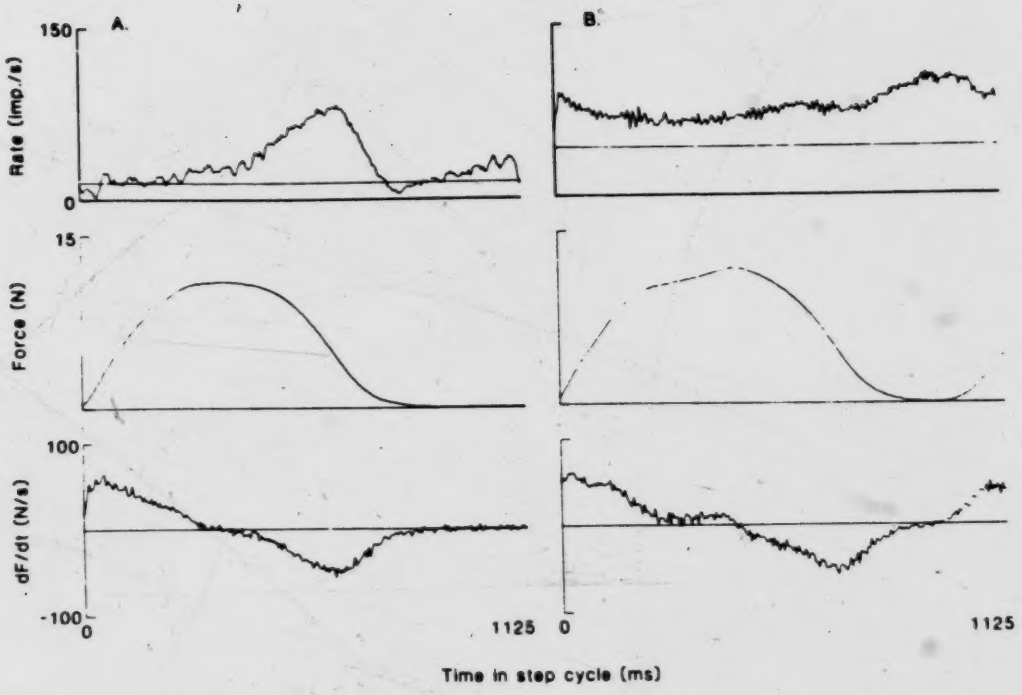
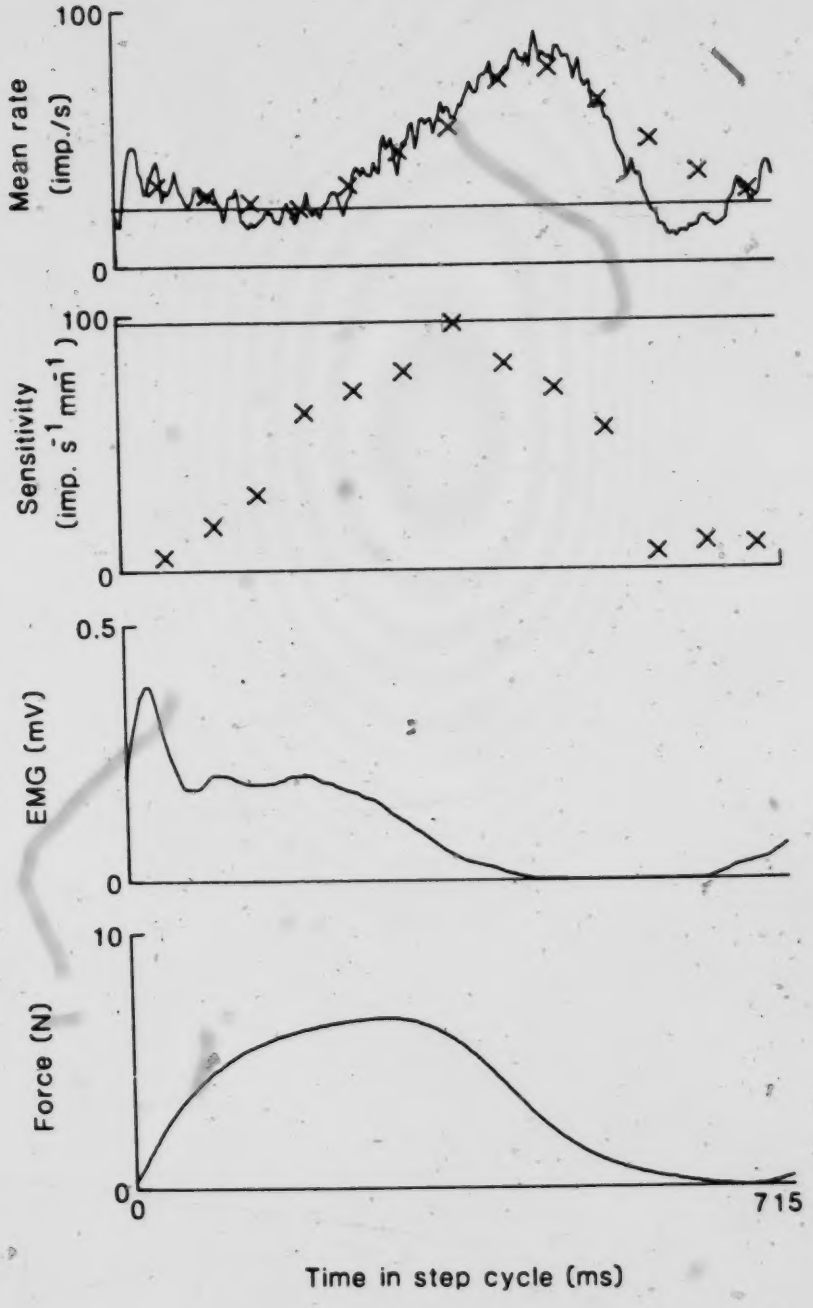


Figure 11. Step cycle histograms for a primary afferent (A) and for a secondary afferent (B). The horizontal line represents the mean discharge rate of the afferent when stretches were applied while the cat was at rest. The afferent activity during the step cycle can be compared with the force and rate of change of force.

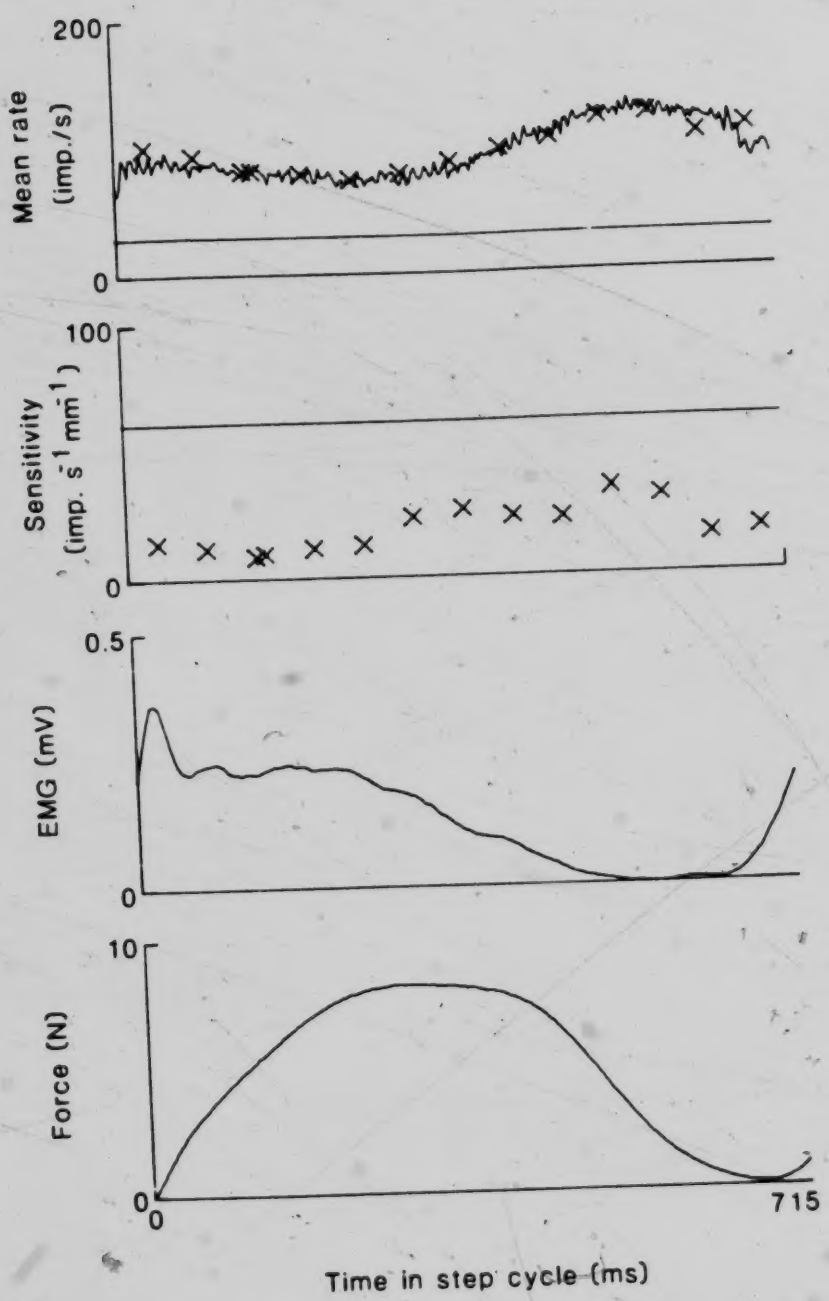
Figure 12. Mean rate, and sensitivity to stretch, of a primary afferent during the step cycle. The mean rate and sensitivity were determined from the average response of the afferent to sinusoidal stretch (4 Hz, 0.31 mm) at each of 14 different times in the step cycle. Although each value (X) pertained to the afferent activity over 250 ms, it was plotted at the time corresponding to the middle of the stretch period. The step cycle histogram has been superimposed on the values for mean rate. The horizontal lines represent the values for mean rate and sensitivity when the cat was at rest. EMG and force are shown for comparison of timing. Because the period shown is less than the duration of the average step, the fourteenth point has been omitted.


Mean rate (imp./s)
100
0
Sensitivity (imp. s⁻¹ mm⁻¹)
100
0
EMG (mV)
0.5
0
Force (N)
10
0
0
715
Time in step cycle (ms)

for a secondary afferent in Fig. 13. Each X represents the sensitivity or mean rate at that particular time in the step cycle. The values at each phase were calculated from the average response to one cycle of stretch, which for 4 Hz stretches covers 250 ms. The values were plotted at the time in the step cycle corresponding to the midpoint of the stretch. For example, the average response to stretches beginning 0-54 ms following the step marker was plotted at 152 ms, corresponding to the midpoint of the first phase, 27 ms, plus the time from the stretch marker to the midpoint of the cycle, 125 ms. Thus the mean rate and sensitivity for those stretches which started in the thirteenth and fourteenth phases were actually plotted near the beginning of the step cycle, since the midpoint of the stretch cycle occurred early in the next step cycle. In the first traces of Figs. 12 and 13 the step cycle histograms have been superimposed on the plotted values of the mean rate. The close temporal agreement indicates that the midpoint of the stretch, rather than elsewhere in the 250 ms time span, was the best time at which to plot the mean and sensitivity values.

The horizontal lines represent the stretch sensitivity or mean impulse rate during the resting state. For the primary afferent illustrated in Fig. 12, the peak sensitivity just exceeds the resting sensitivity. The peak sensitivity was greater than the resting sensitivity for 5 other primary afferents. For the remaining 9 primary

Figure 13. Mean rate, and sensitivity to stretch, of a secondary afferent during the step cycle. Responses to 4 Hz, 0.44 mm sinusoidal stretches were averaged at 14 different times in the step cycle. Each X represents the mean rate or sensitivity at that particular time in the step cycle. The step cycle histogram has been superimposed on the values for mean rate. The horizontal lines represent the values for mean rate and sensitivity when the cat was at rest. EMG and force are shown for comparison of timing.


200
Mean rate
(imp./s)
0
100
Sensitivity
(imp. s^{-1} mm^{-1})
0
0.5
EMG (mV)
0
10
Force (N)
0
0
715
Time in step cycle (ms)

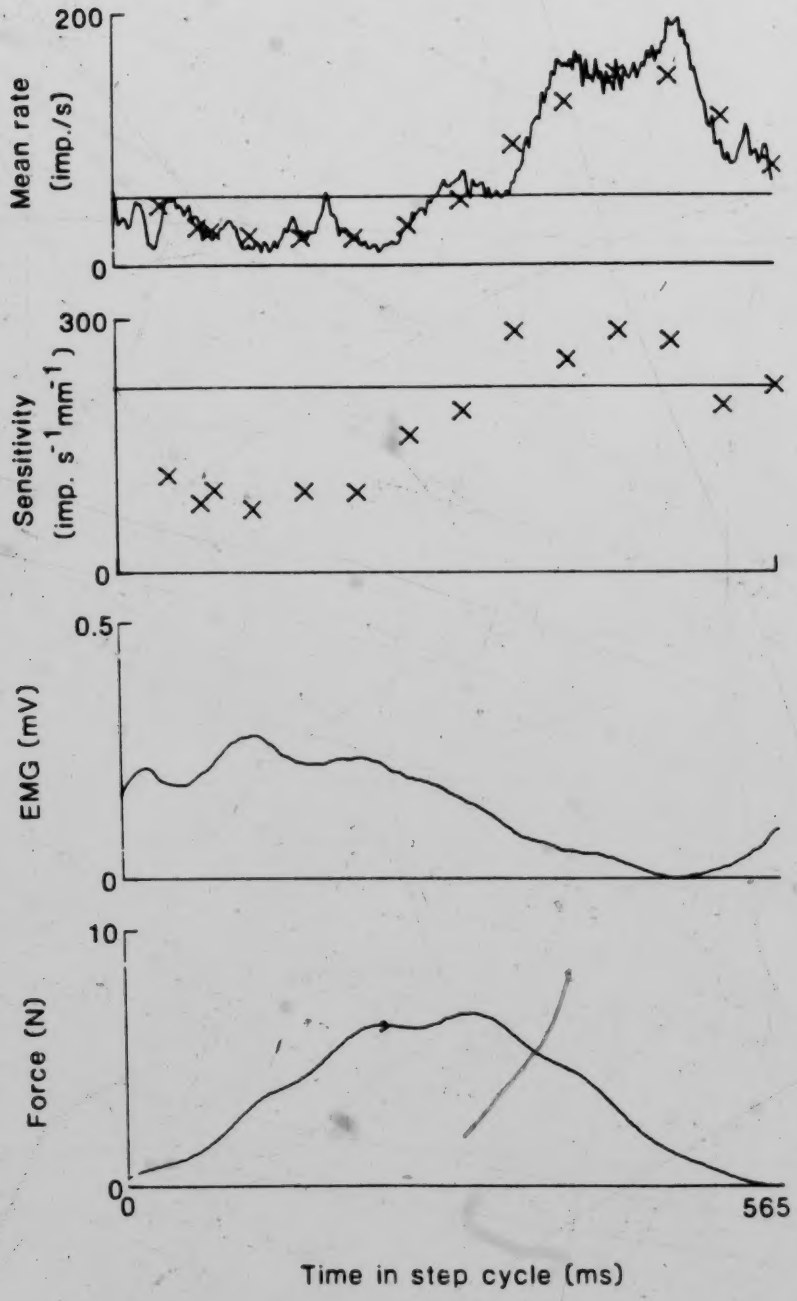
afferents studied with 4 Hz sinusoidal stretch, the sensitivity was below the resting sensitivity at all times in the step cycle. The sensitivity was highly modulated during the step cycle for all the primary afferents studied. The peak sensitivity to 4 Hz sinusoidal stretch occurred on average 18 $\pm$ 18% of the step cycle after the peak in tension. This difference was significant at $p < 0.05$. To account for the modulation in sensitivity, a rhythmically varying pattern of fusimotor activity must have influenced the primary afferents.

For most of the secondary afferents, the sensitivity was decreased below the resting value and was only slightly modulated. This can be accounted for by a relatively steady level of fusimotor activity during walking. The large increase in mean impulse rate during walking is indicative of a high level of fusimotor activity.

Not all the secondary afferents, however, showed the illustrated changes in sensitivity. The peak sensitivity of three afferents was above the resting sensitivity, and during the step cycle the sensitivity of two of these afferents was quite variable.

To improve the time resolution of the step cycle changes, the responses of primary afferents to 10 Hz sinusoidal stretches were also studied. Each value for mean rate and sensitivity was obtained from a span of 100 ms rather than 250 ms. The values were again plotted at the times corresponding to the middle of the stretch. An example is shown in Fig. 14. However, a problem with using

Figure 14. Mean rate, and sensitivity to stretch, of a primary afferent at 14 different times in the step cycle. 10 Hz, 0.26 mm sinusoidal stretches were used. The horizontal lines represent the values for mean rate and sensitivity when the cat was at rest. The EMG and force are shown for comparison.


200
Mean rate
(imp./s)
0
300
Sensitivity
(imp. s^{-1}mm^{-1})
0
0.5
EMG (mV)
0
10
Force (N)
0
0
565
Time in step cycle (ms)

stretches of 10 Hz became apparent. At this frequency the afferent impulses had a tendency to occur only at fairly specific phases of the step cycle. When this phenomenon known as phase-locking occurred, the method of analysis resulted in the amplitude of the fitted sine changing in proportion to the mean rate, at about twice the size. Meaningful information about the fusimotor activity could not then be interpreted from the sensitivity. For this reason, results from 10 Hz stretches applied during walking will not be discussed.

To allow interpretation of the changes in sensitivity and mean rate in terms of fusimotor activity, control experiments were performed on anesthetized cats. The effects of stimulating identified γ-motoneurons on the afferent's response to sinusoidal stretches were analysed. This was necessary because such effects have not been studied for stretches with the parameters we used.

CHAPTER FIVE

STRETCH RESPONSES OF MUSCLE SPINDLE AFFERENTS DURING γ-MOTONEURON STIMULATION

5.1 METHODS

Nine anesthetized cats were used in acute experiments for the investigation of the sinusoidal stretch response of primary afferents when the fusimotor drive to the spindle was controlled. The stretches covered the same range of parameters as those used for the premammillary cats. Identified γ-motoneurons influencing the isolated afferents were stimulated at steady rates and at modulated rates representative of the patterns of activity that have been recorded during locomotion in premammillary cats. These experiments provided a basis for the interpretation of the changes in the stretch response during walking.

The cats were anesthetized with Nembutal at an initial dose of 0.5 ml/kg (0.32 mg sodium pentobarbitol/kg). The surgical procedure was the same as that previously described except for the following differences. The second common carotid artery was not ligated and the animal was not decerebrated. So that the ventral roots could be reached, a wider laminectomy was made. The L_6 to S_2 dorsal and ventral roots were cut close to the spinal cord. The soleus nerve was stimulated to determine from the compound potentials of the L_7 and S_1 roots the easier dorsal and ventral roots for isolating functionally single soleus nerves. (The L_6 and S_2

roots were cut to reduce any possible unwanted reflexes.)

Up to six single soleus primary muscle spindle afferents were dissected from the L_7 or S_1 dorsal roots. They were identified by their conduction velocities, and their responses to twitch contractions and large ramp stretches. To isolate single γ-motoneurons affecting these afferents, the L_7 or S_1 ventral root was divided into sixteen filaments. A filament was stimulated at 100 stim./s and each of the afferents was checked in turn for a fusimotor influence. If an increase in mean rate and a change in the dynamic response to large ramp stretches were found, the soleus nerve and EMG activities were averaged using the stimulation pulses to trigger the sweeps. In this way, the number of motoneurons in the filament was determined. The filament that influenced one or more isolated afferents was divided until there were no EMG potentials, thus eliminating the α -motoneurons to the soleus, and ideally, until only one distinct neural potential, that of a single γ -motoneuron, was observed. Identification of a γ- motoneuron was based on the following criteria: enhancement of the afferent's firing rate, no EMG activity, and conduction velocity less than 55 m/s (Matthews, 1972). Conduction latency was measured from the soleus nerve average. Not infrequently, efferent potentials from γ-motoneurons influencing other soleus afferents, or from γ or α motoneurons supplying other muscles, were present in the average. The stimulation voltage was adjusted so that the potential corresponding to the γ-

motoneuron influencing the isolated afferent of interest could be determined. Sometimes stimulation of a single soleus motoneuron simultaneously produced EMG activity and an increase in the firing rate of the afferent. At low rates of stimulation, unfused contractions from α-motoneuron activation of an extrafusal fiber in series with the spindle could produce this effect as could activation of a β-motoneuron. The identification of a β-motoneuron would only have been made if the increased bias of the afferent was maintained at high rates of stimulation.

In a few experiments, pairs of single γ-motoneurons influencing the same afferent were looked for. The effect of each of the sixteen filaments on an afferent was checked in turn, and then the relevant filaments were divided until two γ-motoneurons influencing the afferent were isolated.

Stimulation of the ventral root filament at 100 stim./s was used to classify a γ-motoneuron as dynamic or static according to the change in the dynamic response of the afferent to large ramp stretches of 4 mm at 8 mm/s.

The afferent response to sinusoidal stretch was studied under different states of fusimotor activation. The frequencies of stretch were 4 and 10 Hz, and the amplitudes were 0.05, 0.2, and 0.7 mm, thus including the parameters used for the premammillary cat. The identified γ-motoneuron was stimulated at steady rates of usually 10, 20, 50, 100, and 160 stim./s. Although γ-motoneurons firing at 160 imp./s have not been observed during locomotion, this rate

was used to represent the possible net effect of two or more γ-motoneurons influencing the afferent simultaneously. Stretch cycle histograms were computed for each of these conditions, and for the condition of no fusimotor activity.

To simulate the phasically modulated pattern of fusimotor activity that has been recorded during locomotion, the isolated γ-motoneuron was also stimulated with a modulated rate which varied between 10 and 70 stim./s over a period of 660 ms. An electronic neural analog, influenced by a 1.5 Hz sinusoidal input, was used to generate this stimulation pattern. The analysis was analogous to that used to determine the stretch response at different times in the step cycle. A marker for the 1.5 Hz sinusoid was used instead of the step cycle marker. The stretch responses were determined at fourteen phases of the 660 ms period.

When two γ-motoneurons influencing the primary afferent were isolated, one of them was stimulated with steady rates, while the other was stimulated with the modulated pattern.

5.2 RESULTS

5.2.1 TONIC STIMULATION

The effect of stimulating a γ-motoneuron at 100 stim./s on the response of a primary afferent to 4 Hz, 0.2 mm sinusoidal stretch is shown in Fig. 15 for a dynamic and a static γ-motoneuron. The mean rate was increased 19 imp./s by the γ_D action, and 55 imp./s by the γ_S action. The sensitivity was increased 74% by the action of the dynamic

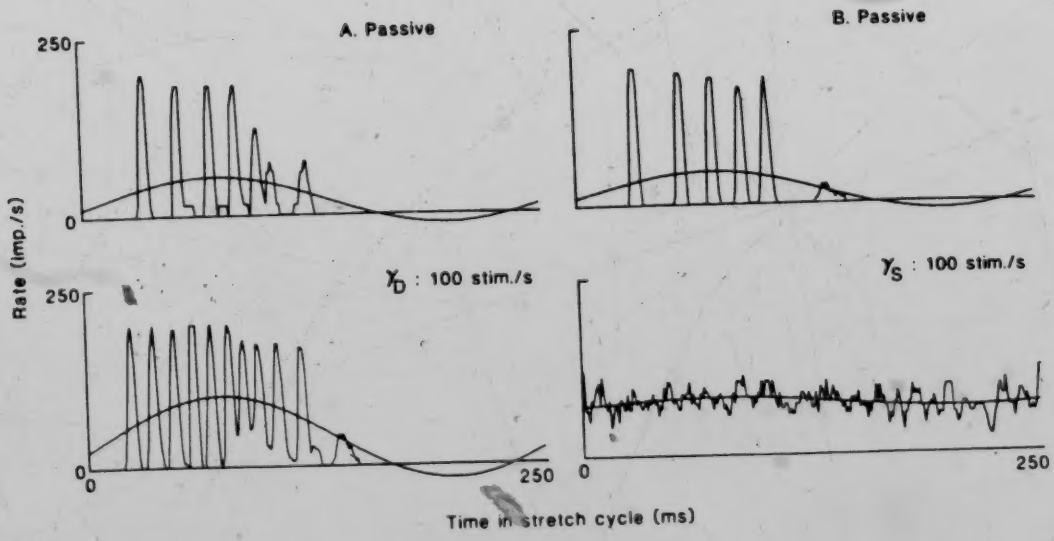


Figure 15. The effect of γ-motoneuron stimulation on the response of a primary afferent to 4 Hz, 0.2 mm sinusoidal muscle stretch. A. The mean discharge rate and the modulation of the rate were increased by dynamic γ-motoneuron stimulation at 100 stim./s. B. The mean rate was increased and the modulation of the rate was decreased by static γ-motoneuron stimulation at 100 stim./s.

γ-motoneuron, and was decreased 71% by the action of the static γ-motoneuron.

The afferent response was generally graded with the rate of stimulation. This is shown in Fig. 16 for the primary afferent of Fig. 15. The points were fitted with the best straight line using the method of least mean squares. The slopes of the lines represent the change in the number of impulses for each stimulation pulse to the γ-motoneuron, or the % change in sensitivity for each stimulation pulse per sec. From the values of the slopes in Fig. 16, the mean rate was increased 0.19 imp./stim. with dynamic γ-motoneuron stimulation, and 0.58 imp./stim. with static γ-motoneuron stimulation. Dynamic γ- motoneuron caused an increase in sensitivity by 0.78% stim.$^{-1}$ s^{-1}, whereas static γ-motoneuron stimulation caused a decrease by 0.80% stim.$^{-1}$ s^{-1}. On average, for the primary afferents studied, γ_D stimulation produced a change in the mean rate by 0.32 imp./stim., and a % change in the sensitivity by 1% stim.$^{-1}$ s^{-1} in response to 4 Hz, 0.2 mm sinusoidal stretch. γ_S stimulation produced a change in the mean rate by 0.42 imp./stim., and a % change in the sensitivity by -0.25% stim.$^{-1}$ s^{-1}. The average values of the slopes, which were also determined for 0.05 and 0.7 mm, 4 Hz stretches, and for 0.05, 0.2, and 0.7 mm, 10 Hz stretches, are shown in Table 1.

The afferent response to static γ-motoneuron stimulation was not consistent. For example, driving was elicited by two of the static γ-motoneurons. An afferent

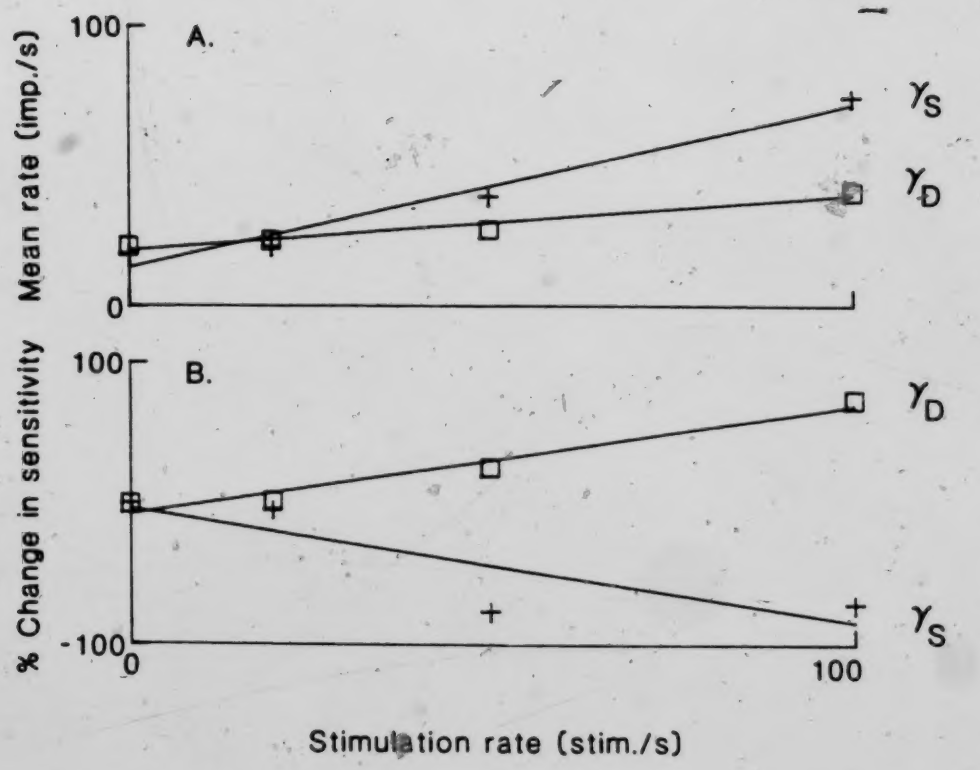


Figure 16. The effect of increasing the rate of γ-motoneuron stimulation on the primary afferent response to sinusoidal stretch. The points were fitted with a straight line using the method of least mean squares. The mean discharge rate of the afferent (A) increased with increases in the stimulation rate of both the dynamic (□) and the static (+) γ-motoneuron, but the slope is greater for the static γ-motoneuron stimulation. The afferent sensitivity (B) increased with increases in the rate of stimulation of the dynamic γ-motoneuron (□), but decreased with increasing stimulation of the static γ-motoneuron (+).

Freq. (Hz)		4			10		
	Amp. (mm)	0.05	0.2	0.7	0.05	0.2	0.7
γ_D	Δ in mean rate (imp./s)	0.23	0.32	0.67	0.23	0.42	0.95
	Δ in sensitivity (%Δ sens./stim./s)	1.02	1.00	1.78	0.74	0.86	1.67
	no. of afferents	5	8	3	5	8	3
γ_S	Δ in mean rate (imp./s)	0.43	0.42	0.44	0.34	0.32	0.32
	Δ in sensitivity (%Δ sens./stim./s)	-0.53	-0.25	-0.07	-0.32	-0.11	0.04
	no. of afferents	8	11	3	8	11	3

Table 1. Effects of dynamic and static γ-motoneuron stimulation on the primary afferent stretch response. Values were computed from slopes of straight lines, such as those shown in Fig. 16, and averaged. The values represent the average change in impulse rate or % change in sensitivity for each extra stim./s.

impulse was produced for each stimulation pulse up to 100 stim./s. The stretch sensitivity was therefore low at all rates of γ-motoneuron stimulation.

Other exceptions to the graded decrease in sensitivity with increasing rates of static γ -motoneuron stimulation were observed in that the sensitivity sometimes actually increased with low rates of stimulation and then decreased when the rate of stimulation reached 50 or 100 stim./s. This can be explained by the afferent impulses not occurring throughout the stretch cycle, but being phase locked to the stretch. In Fig. 15, this is apparent for the condition of no fusimotor activity. At low rates of static γ-motoneuron stimulation the afferent increased firing but sometimes still only fired during the stretch phase. As well as an increased mean rate this behaviour resulted in an increased modulation and hence increased sensitivity. Not until the afferent fired throughout the stretch cycle did the sensitivity decrease.

Phase locking was greatest for the 0.7 mm stretches and least for the 0.05 mm stretches. Small amplitude (0.05 mm) stretches, to which we found an opposite effect of static and dynamic fusimotor stimulation on the afferent sensitivity, could not be used in the premammillary cat experiments because the stretch responses would not be discernible in the locomotor activity.

Because the afferent is considerably more disposed to phase locking with 10 Hz than 4 Hz stretches, the static

action is not clearly distinct from the dynamic action during 10 Hz stretches, even at 0.2 mm. Thus the application of 10 Hz stretches during locomotion would not be useful in determining the predominant type of fusimotor activity.

The response of one secondary afferent to 4 Hz, 0.2 mm sinusoidal stretch was also recorded. Stimulation of a static γ-motoneuron at 100 stim./s caused an increase in the mean rate by 25 imp./s and a decrease in the sensitivity by 53%.

5.2.2 MODULATED STIMULATION

The effect of stimulating a γ-motoneuron with a modulated rate, representative of the activity in phasically modulated gamma axons during walking, was analyzed. The response of a primary afferent to 0.2 mm stretch during dynamic γ-motoneuron stimulation is shown in Fig. 17A, and during static γ-motoneuron stimulation is shown in Fig. 17B. The rate of stimulation varied between 10 and 70 stim./s over a period of 660 ms. The increase in rate of the γ_D motoneuron was followed by increases in both the mean rate and sensitivity whereas the increase in stimulation rate of the γ_S motoneuron was followed by an increase in the mean rate and a decrease in the sensitivity. The afferent activity, averaged using a 1.5 Hz marker, generally reached a peak about 80 ms following peak γ_D stimulation, or about 65 ms following peak γ_S stimulation. This lag was due to conduction and synaptic delays and to the contraction time

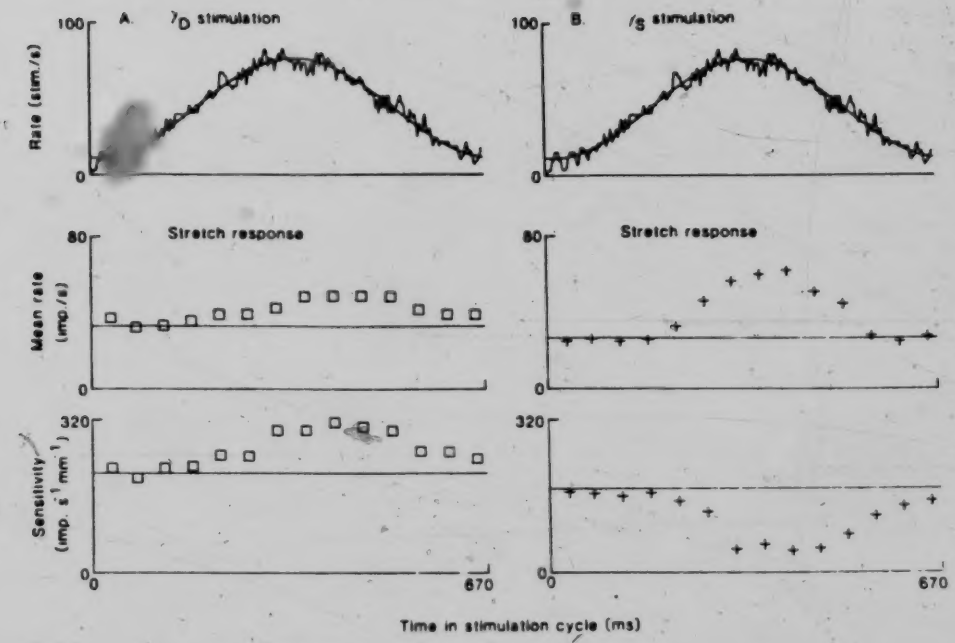


Figure 17. Mean rate, and sensitivity to stretch, of a primary afferent during γ-motoneuron stimulation at a modulated rate. The stimulation rate was varied between 10 and 70 stim./s over a period of 660 ms, and is illustrated as a smoothed histogram with a fitted curve. During modulated γ_D stimulation (A), the mean discharge rate and sensitivity followed the change in rate of stimulation with a lag. During γ_S stimulation (B), the mean discharge rate increased and the sensitivity decreased as the stimulation rate increased. The horizontal lines represent the values for mean rate and sensitivity for the passive ending.

of the intrafusal fibers. However, because each value was calculated from a 100 or 250 ms period (depending on whether 10 Hz or 4 Hz stretches were applied), the timing of the values for rate and sensitivity, relative to the phase of the gamma axon stimulation, was imprecise. This point must be considered when interpreting the timing of the changes in sensitivity that occur during the step cycle.

5.2.3 COMBINED STIMULATION

As well as a phasically modulated pattern, a tonically modulated pattern of fusimotor activity has been recorded during walking. A muscle spindle is quite probably influenced simultaneously by both activity patterns. To determine the primary afferent response under such a combined influence, a dynamic γ- motoneuron was stimulated with one pattern while a static γ-motoneuron to the same muscle spindle was stimulated with the other pattern. In Fig. 18A, stimulation of the γ_S axon at a steady rate of 100 stim./s resulted in an increased mean rate of discharge, but decreased modulation, to the applied length changes. When stimulation of the γ_D axon with a modulated rate was added, the mean discharge rate was increased, but the amplitude of the stretch response varied above and below the amplitude when there was no fusimotor activity. In Fig. 18B, the γ_D axon was stimulated at a steady rate of 100 stim./s and the γ_S axon was stimulated with the modulated rate. This resulted in an increase in the mean discharge rate, but the amplitude of the stretch response, although variable, was

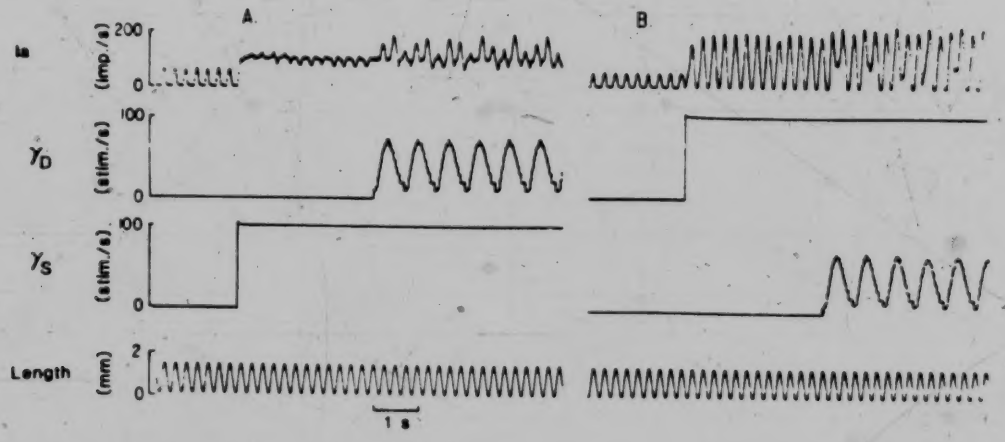


Figure 18. The response of a primary afferent to sinusoidal stretch (4 Hz, 0.7 mm) during combined stimulation of a γ_S and a γ_D. One γ-motoneuron was stimulated at a tonic rate of 100 stim./s and the other was stimulated at a modulated rate of 40 $\pm$ 30 stim./s. The pulse train, representing the firing of the afferent, was low pass filtered to convert it to a rate of firing and then was printed with a pen recorder. The stimulation rates of the γ-motoneurons were converted to instantaneous frequencies by an interspike interval converter and were printed in the same manner. Tonic stimulation of a γ_S, with modulated stimulation of a γ_D (A), resulted in an afferent response similar to that which occurred during walking. Tonic stimulation of a γ_D, with modulated stimulation of a γ_S (B), resulted in a large modulation in the afferent firing rate, which varied out of phase with the modulated stimulation.

always greater than that which occurred in the passive spindle.

CHAPTER SIX

DISCUSSION

6.1 FUSIMOTOR ACTIONS ON THE AFFERENT RESPONSE TO STRETCH

Muscle length changes and fusimotor activity interact to determine the activity in muscle spindle afferents. The actions of the γ-motoneurons are dependent on the size and speed of the movement. For the premammillary cats, we mainly used stretches of 4 Hz, 0.2 mm. Using these same parameters of stretch in control experiments, stimulation of γ-motoneurons of either type increased the mean rate of firing of primary afferents. The stretch sensitivity of primary afferents was increased by γ_D stimulation at 100 stim./s and decreased by γ_S stimulation. At lower rates of stimulation, some static γ-motoneurons increased the sensitivity. Such paradoxical effects of static γ-motoneurons have also been noted by others (Hulliger, 1984). In part, this effect can be attributed to phase-locking of the afferent impulses. The effect of static γ-motoneuron stimulation on a secondary afferent was to increase the mean rate and decrease the sensitivity.

We also investigated the response of afferents to gamma nerve fiber stimulation, using other parameters of stretch. Although we found that the difference in the effects of static and dynamic γ-motoneuron stimulation on primary afferents was most apparent with 4 Hz stretches of smaller amplitude, had they been used during locomotion, the

responses to these stretches would have been over-ridden. Phase-locking became worse as the frequency or amplitude of the stretch was increased and was quite severe for 10 Hz, 0.7 mm stretches. For these reasons, our analysis of the primary afferent response during walking was mostly limited to 4 Hz stretches between 0.2 and 0.5 mm. Knowing how fusimotor activity influences the afferent stretch response allows the underlying fusimotor activity to be deduced from the stretch responses during locomotion.

6.2 DEDUCTION OF FUSIMOTOR ACTIVITY DURING LOCOMOTION FROM AFFERENT STRETCH RESPONSES

6.2.1 SECONDARY AFFERENT STRETCH RESPONSES

Secondary afferents are generally only affected by static γ-motoneurons, so that the increased mean rate and decreased sensitivity to sinusoidal stretches during the walking period can be explained by an increase in static γ-motoneuron activity. Similarly, an increase in static γ-motoneuron activity can explain the increased mean rate and decreased dynamic sensitivity in response to ramp stretches during walking as compared to during quiet standing.

The strength of the increased static γ-motoneuron activity can be estimated from the afferent's sensitivity, the amount of internal shortening, and the change in mean rate, in conjunction with a value for the effect of γ_S stimulation on increasing the rate. As an example, the average sensitivity of one secondary afferent during walking

was about 16 imp. s^{-1} mm^{-1}. The calculated internal shortening of the muscle was 3.4 mm, so that in response to this alone, the afferent would have decreased firing by about 54 imp./s. However, the firing rate increased by 25 imp./s on average for this period. Thus the static γ-motoneuron activity was sufficiently strong to cause an overall change in the afferent firing rate by 79 imp./s.

In our control experiments, using stretches of the same parameters, a secondary afferent increased its firing rate by 24 imp./s in response to static γ-motoneuron stimulation at 100 stim./s, i.e., 0.24 imp./stim. However, this was the only secondary afferent that we studied. Others (Cussons et al, 1977), found that the mean discharge rate during 1 Hz stretching increased by about 30 imp./s for an increase in the rate of γ_S stimulation from 50 to 100 stim./s: 0.6 imp./stim. This agrees well with an average increase in the secondary afferent rate of 74 imp./s by γ_S stimulation at 120 stim./s, i.e., 0.6 imp./stim., with the muscle at constant length (Andersson et al, 1968). It is prudent to use this latter value for the purpose of approximating static γ-motoneuron strength. That the experimental conditions were different should not matter, especially considering the similarity of the two published results.

Thus an increase in the firing rate of the secondary afferent by 79 imp./s would require an increase by 132 imp./s of the static γ-motoneuron activity affecting it. Although this is just one example, it is useful as a general indication of the strength of the static action. Naturally,

variability in the strength can be expected due to differences among spindles in the static γ-motoneuron innervation.

As evidenced by the maintained reduction in stretch sensitivity throughout all phases of the step cycle for most of the secondary afferents, such strong γ_S activity was relatively tonic.

In a few secondary afferents, the sensitivity varied somewhat during the step cycle. This could be due to the analysis procedure not providing an accurate description of the true state, as would result from poor correlation of the fitted sinusoids to the afferent activity. This was most evident when only a small number of steps were averaged or when the step length varied greatly. The different effects of static bag_2 and chain fiber contraction on stretch sensitivity, and their particular predominance on the secondary afferent activity in different spindles, may also contribute to the variation in results.

6.2.2 PRIMARY AFFERENT STRETCH RESPONSES

Primary afferents are influenced by both static and dynamic γ-motoneurons. During the step cycle, the sensitivity was greatly modulated. Phasically modulated activity in dynamic γ-motoneurons accompanying the tonic activity in static γ-motoneurons could account for this. The sensitivity was highest slightly after the time of peak force and was low in the absence of EMG activity. In a freely moving cat, this would correspond to high sensitivity

during stance and low sensitivity during the swing phase. (The imprecision involved in matching the calculated sensitivity to a particular phase of the step cycle does not allow interpretation of the timing to be more specific.)

In the anesthetized cats, peak sensitivity occurred about 100 ms after the peak γ_D stimulation rate. To cause the high sensitivity at the time of high force in the step cycle, the dynamic γ-motoneurons must have been most active during the time the force was increasing.

The tonic static γ-motoneuron activity would have reduced the primary afferent sensitivity from that which would have been set by activity in dynamic γ-motoneurons alone. The dynamic responsiveness to ramp stretches and the sensitivity to sinusoidal stretches were reduced on average during the walking period compared to during quiet standing.

6.3 IDENTIFICATION OF PHASIC AND TONIC γ-AXONS AS DYNAMIC AND STATIC γ-MOTONEURONS

In other experiments using premammillary cats, gamma nerve fibers from soleus were named according to the activity pattern recorded from them during walking (Murphy et al, 1984). Tonic γ-axons (γ_t) had a low resting rate (< 20 imp./s), but fired at a high rate (> 50 imp./s) during walking. During the step cycle, the rate was only modulated $\pm$ 9 imp./s and was greatest just before the peak EMG. Phasic γ-axons (γ_p) fired at more than 50 imp./s when the cat was resting. During walking, the rate slightly

decreased on average and was highly modulated (± 23 imp./s) with the peak activity following the peak EMG, at about the time of peak tension.

6.3.1 EVIDENCE FOR γ_p'S AS DYNAMIC γ-MOTONEURONS

The modulation during walking in primary afferent sensitivity, but not in secondary afferent sensitivity, indicates that the phasic pattern of activity probably occurred in dynamic γ-motoneurons. The peak activity in the γ_p axons is at the appropriate time to account for the peak sensitivity of the primary afferents. The sensitivity during quiet standing, which exceeded the sensitivity during most of the step cycle, is in agreement with the high resting rate of γ_p axons occurring in dynamic γ-motoneurons.

In an anesthetized cat, stimulation of a dynamic γ-motoneuron at a modulated rate, together with stimulation of a static γ-motoneuron at a tonic rate, caused a modulation in the amplitude of the stretch response of a primary afferent, as shown in Fig. 18A. The amplitude of the response was at times below, and at other times greater, than that of the passive ending. This observation is consistent with the modulation in primary afferent sensitivity which occurred during walking. Conversely, when the static γ-motoneuron was stimulated with a modulated rate and the dynamic γ-motoneuron with a tonic rate, the sensitivity remained high on average, with the peak sensitivity out of phase with the time of peak stimulation, as shown in Fig. 18B. The static γ-motoneuron was not

activated in this way during walking since the peak γ_p activity would have caused the sensitivity to be low at the time of peak tension.

6.3.2 EVIDENCE FOR γ_t'S AS STATIC γ-MOTONEURONS

The maintained reduction in secondary afferent sensitivity during walking suggests that the tonic pattern of activity probably occurred in static γ-motoneurons. The sensitivity of the secondary afferent was always lowest near the beginning of the EMG: at the time of peak γ_t activity. Furthermore, a second peak in impulse rate, shown by some primary afferents, occurred at this time too. This is when internal length changes would otherwise tend to unload the spindle. Static γ-motoneurons are more effective at biasing the spindle and preventing spindle unloading than are dynamic γ-motoneurons. During walking, the estimated total increase in static γ-motoneuron activity influencing a secondary afferent, when divided by several motoneurons, can be explained by the increase in mean rate of γ_t axons but not of γ_p axons. Another reason why the γ_t axons are unlikely to belong to dynamic γ-motoneurons is because the peak primary afferent sensitivity did not follow the peak γ_t activity.

The conclusion that γ_t axons belong to static γ-motoneurons and γ_p axons belong to dynamic γ-motoneurons, confirms the identification made earlier (Murphy et al, 1984), which was based mainly on the resting rates of a small number of functionally classified γ-motoneurons

maintained in continuity.

6.4 FUNCTIONAL IMPLICATIONS

6.4.1 PRIMARY AFFERENTS

As mentioned previously, the precise timing of the sensitivity changes, relative to EMG and force levels, is equivocal. However, it is clear that the sensitivity of the primary afferent is relatively high during the latter three-quarters of force development and is low in the absence of active force. In the freely moving cat, this would correspond to relatively high sensitivity during the stance phase and low sensitivity during the swing phase.

The low sensitivity would limit the afferent response as the muscle is passively stretched during flexion. At a walking speed of less than 3 km/h the muscle is lengthened by more than 10 mm at a rate of about 100 mm/s (Goslow et al, 1973), so that a passive ending would fire rapidly during this period. The low sensitivity thus aids in preventing reflex activity from interfering with the movement. Without stretch-induced co-activation, the movement during the swing phase can be achieved with less muscle activity for any desired speed and thus allow the animal to operate with greater efficiency.

During the stance phase, reflex activity in soleus reinforces the ongoing movement rather than opposes it as happens in the swing phase. It is appropriate, therefore, for the spindle sensitivity to be increased during extension

by the addition of dynamic γ-motoneuron activity. The consequent burst of afferent firing when the active muscle is stretched briefly by about 3 mm upon foot contact (Goslow et al, 1973) reflexly increases extensor activity, resulting in a greater propulsive force to accelerate the animal up and forward. In a de-afferented cat, the force of each step is indeed reduced (Wetzel et al, 1976). The relatively high sensitivity is sustained during the period of active force development. Thus, any perturbation encountered within the stance phase, even as the weight is transferred forward and the active muscle shortens, will result in reflex activity to counteract the perturbation. An example is an unanticipated change in the terrain. When an animal's foot lands on an uphill slope, the ankle extensors are longer and because of the extra load, shorten more slowly. The resulting increased afferent activity leads to continued extensor support and propulsion. The reflex force has a delay of about 40 ms (Grillner, 1972) and so can contribute to the support phase extension during walking, since this phase lasts about 400 ms (Goslow et al, 1973).

The changing sensitivity to stretch during the step cycle works together with the locomotor drive potential to determine the reflex response. In flexion, the membrane potential of a soleus α-motoneuron is more negative, and thus is less excitable, than in extension (Shefchyk et al, 1984). A given rate of afferent impulses will not achieve the generation of as many action potentials in the soleus α-

motoneurons during flexion. The low sensitivity at this time means the change in afferent firing rate for any length change will be less. Thus the two mechanisms, one which reduces the peripheral input to the α-motoneuron, the other which reduces the output, act co-operatively to minimize opposition to movement during the swing phase. Conversely, the high stretch sensitivity and less negative membrane potential during extension together increase the reflex response and enable the movement to be reinforced when perturbed.

Because the dynamic γ-motoneuron activity is accompanied by static γ-motoneuron activity, the sensitivity to 0.2 mm, 4 Hz stretches is not as high as if the dynamic γ-motoneuron activity had occurred alone (Hulliger et al, 1977b). Furthermore, the sensitivity to very small stretches would be low: equivalent to that during solely static γ-motoneuron activity (Hulliger et al, 1977b). Perhaps a high sensitivity to small stretches is not important during locomotion: very small perturbations are unlikely to disturb the stepping pattern. This is in contrast to the condition of quiet standing. In this situation, a corrective response made quickly to a tiny length change before a larger error develops is important in the maintenance of posture. The high dynamic sensitivity to small stretches required for this can not be achieved by a spindle under the influence of static γ-motoneuron activity. The very low firing rate of presumed static γ-motoneurons recorded in that situation (< 20 imp./s) (Murphy et al,

1984) is in agreement with this function.

6.4.2 SECONDARY AFFERENTS

Static γ-motoneuron activity during the stance phase is not of obvious benefit via the primary afferent. Although the afferent may be prevented from shutting off during slow muscle shortening, the significance of this is not clear. However, static γ-motoneuron activity at this time, and throughout the whole of the step cycle, may be important for the role of the secondary afferent in the control of locomotion. Input from the secondary afferent may be used as a monitor of muscle length. Static γ-motoneuron activity extends the range over which the secondary afferent could function in this way. This is important because, during the normal step cycle, the muscle undergoes quite extensive excursions in length: a shortening to about 6 mm less than the standing length is followed by a total lengthening of about 12 mm (Goslow et al, 1973). Low sensitivity would prevent the secondary afferent from shutting off at short lengths and from firing maximally at increases of only intermediate size. A spindle secondary afferent recording from a freely walking cat has shown maintained firing throughout the step cycle, proportional to muscle length (Loeb & Duysens, 1979), which is consistent with this proposed function.

Because muscle properties can vary with time, such as in fatigue, the same motor command will not always produce the same muscle contraction. Thus, continuous feedback

about muscle length may be important to keep the central nervous system informed about the progress of a movement, so that a corrective action can be applied when necessary to produce the desired movement (Matthews, 1972).

CHAPTER SEVEN

CONCLUSIONS

The firing of spindle afferents is influenced by fusimotor activity and by changes in muscle length. Locomotion in intact cats is not an example of a servo-assisted movement in which α-γ coactivation maintains the afferent activity at an approximately constant level during muscle contraction under normal conditions. Instead, the afferent response is dominated by the length changes. Fusimotor activity would affect the sensitivity to such length changes and also to any disturbances of the normal step cycle.

The intention of this study was to investigate the stretch sensitivity of spindle afferents from soleus during locomotion. Cats decerebrated at the premammillary level walked with three legs on a treadmill while the fourth leg was held isometric except for controlled stretches applied to the soleus muscle. By the choice of appropriate stretch parameters, it was possible to infer the activities in static and dynamic γ-motoneurons in this reduced preparation. These were found to support previous conclusions from direct γ-motoneuron recordings: tonic activity in static γ-motoneurons and phasic activity in dynamic γ-motoneurons. The interaction of their combined effects on spindle afferent sensitivity was determined.

Although it is interesting to interpret the results in terms of normal walking in the intact cat, it must be

remembered that fusimotor activity in the premammillary cat could differ from that in the intact cat. However, afferent recordings from intact and mesencephalic cats during walking show general similarities; the discrepancies can be explained by differences in the strength of the fusimotor activity rather than by differences in the timing (Prochazka et al, 1976; 1977).

The sensitivity of primary afferents was modulated during the step cycle. It was high during the period of active force development which corresponds to the support phase of the step cycle in a freely walking cat. High sensitivity at this time contributes, along with the locomotor drive potential, to increasing the reflex response and enabling the extensor activity to be reinforced. The sensitivity was low in the absence of force, and by contributing to a decrease in reflex response it would prevent opposition of the limb movement during the swing phase. On average, the sensitivity was reduced relative to that apparent in quiet standing. Maintenance of posture is improved by high sensitivity. The tonic low sensitivity of the secondary afferent during walking may be important to extend the range over which the afferent can provide meaningful input about muscle length.

This interpretation applies to muscle spindle afferents from an ankle extensor in the control of walking. It would be interesting to know how generally it applies: whether for other speeds, other muscles or even other rhythmic

movements. With increases in speed the average length of the muscle increases and the amount of active lengthening associated with the E2 phase increases. However, the shorter period of the step cycle may cause reflex effects to occur inappropriately. Flexor muscles have quite a different role in locomotion than extensors so the fusimotor activity and afferent stretch sensitivity may reflect this difference. The bifunctional muscles of the hip and knee, and the muscles of the forelimb, can be expected to be controlled differently as well. The results of initial investigations (Cabelguen, 1981; Cabelguen et al, 1984) do suggest the utilization of other strategies. The muscle spindle function in the control of other rhythmic movements, such as breathing and scratching, has yet to be firmly established. The potential for a range of functions is provided by the independent control of static and dynamic γ-motoneurons.

REFERENCES

Akazawa, K., Aldridge, J.W., Steeves, J.D. & Stein, R.B. (1982). Modulation of stretch reflexes during locomotion in the mesencephalic cat. J. Physiol. 329, 553-567.

Andersson, B.F., Lennerstrand, G. & Thoden, U. (1968). Response characteristics of muscle spindle endings at constant length to variations in fusimotor activation. Acta Physiol. Scand. 74, 301-318.

Andersson, O., Forssberg, H., Grillner, S. & Lindquist, M. (1978). Phasic gain control of the transmission in cutaneous reflex pathways to motoneurones during "fictive" locomotion. Brain Res. 149, 503-507.

Andersson, O., Forssberg, H., Grillner, S. & Wallen, P. (1981). Peripheral feedback mechanisms acting on the central pattern generators for locomotion in fish and cat. Can. J. Physiol. Pharmacol. 59, 713-726.

Appenteng, K., Hulliger, M., Prochazka, A. & Zangger, P. (1983). Fusimotor action during movement, deduced by matching the patterns of afferent discharge in freely moving and anaesthetized cats. J. Physiol. 334, 29-30P.

Banks, R.W., Barker, D. & Stacey, M.J. (1981). Structural aspects of fusimotor effects on spindle sensitivity. In Muscle Receptors and Movement, ed. Taylor, A. & Prochazka, A., pp. 5-16. London: MacMillan.

Barker, D. (1983). The current wiring diagram of the muscle spindle. Proc. Int. Union Physiol. Sci. 15, 11.

Boyd, I.A. (1980). The isolated mammalian muscle spindle. Trends Neurosci. 3, 258-265.

Boyd, I.A. (1981a). The muscle spindle controversy. Sci. Prog. 67, 205-221.

Boyd, I.A. (1981b). The action of the three types of intrafusal fibre in isolated cat muscle spindles on the dynamic and length sensitivities of primary and secondary sensory endings. In Muscle Receptors and Movement, ed. Taylor, A. & Prochazka, A., pp. 17-32. London: MacMillan.

Boyd, I.A. (1983). Intrafusal effects of fusimotor stimulation. Proc. Int. Union Physiol. Sci. 15, 11.

Boyd, I.A., Gladden, M.H. & Ward, J. (1983). Two types of static γ-axon having a predominantly chain fibre action in the several cat muscle spindles they supply. J. Physiol. 343, 110P.

Brown, M.C., Crowe, A. & Matthews, P.B.C. (1965). Observations on the fusimotor fibres of the tibialis posterior muscle of the cat. J. Physiol. 177, 140-159.

Cabelguen, J.M. (1981). Static and dynamic fusimotor controls in various hindlimb muscles during locomotor activity in the decorticate cat. Brain Res. 213, 83-97.

Cabelguen, J.M., Orsal, D. & Perret, C. (1984). Discharges of forelimb spindle primary afferents during locomotor activity in the decorticate cat. Brain Res. 306, 359-364.

Crowe, A. & Matthews, P.B.C. (1964). The effects of stimulation of static and dynamic fusimotor fibres on the

response to stretching of the primary endings of muscle spindles. J. Physiol. 174, 109-131.

Cussons, P.D., Hulliger, M. & Matthews, P.B.C. (1977). Effects of fusimotor stimulation on the response of the secondary ending of the muscle spindle to sinusoidal stretching. J. Physiol. 270, 835-850.

Duysens, J. & Pearson, K.G. (1976). Ipsilateral extensor reflexes and cat locomotion. In Neural Control of Locomotion, ed. Herman, R.M., Grillner, S., Stein, P.S.G. & Stuart, D.G., pp. 763-765. New York: Plenum Press.

Engberg, I. & Lundberg, A. (1969). An electromyographic analysis of muscular activity in the hindlimb of the cat during unrestrained locomotion. Acta Physiol. Scand. 75, 614-630.

Forssberg, H. (1979). Stumbling corrective reaction: a phase dependent compensatory reaction during locomotion. J. Neurophysiol. 42, 936-953.

Franz, D.N. & Iggo, A. (1968). Conduction failure in myelinated and non-myelinated axons at low temperatures. J. Physiol. 199, 315-345.

Goodwin, G.M., Hulliger, M. & Matthews, P.B.C. (1975). The effects of fusimotor stimulation during small amplitude stretching on the frequency-response of the primary ending of the mammalian muscle spindle. J. Physiol. 253, 175-206.

Goslow, G.E., Reinking, R.M. & Stuart, D.G. (1973). The cat step cycle. Hindlimb joint angles and muscle length during unrestrained locomotion. J. Morphol. 141, 1-41.

Gottlieb, G.L. & Agarwal, G.C. (1970). Filtering of

electromyographic signals. Am. J. Phys. Med. 49, 142-146.

Grillner, S. (1972). The role of muscle stiffness in meeting the changing postural and locomotor requirements for force development by the ankle extensors. Acta Physiol. Scand. 86, 92-108.

Grillner, S. (1975). Locomotion in vertebrates: central mechanisms and reflex interaction. Physiol. Rev. 55, 247-299.

Grillner, S. (1979). Interaction between central and peripheral mechanisms in the control of locomotion. In Reflex Control of Posture and Movement, ed. Granit, R. & Pompeiano, O., pp. 227-235. Amsterdam: Elsevier.

Grillner, S. (1981). Control of locomotion in bipeds, tetrapods, and fish. In Handbook of Physiology. Sect. 1: The Nervous System, vol. II, pt. 2, ed. Brooks, V.B., pp. 1179-1236. Bethesda, MD: Am. Physiol. Soc.

Grillner, S. & Rossignol, S. (1978). On the initiation of the swing phase of locomotion in chronic spinal cats. Brain Res. 146, 269-277.

Grillner, S. & Zangger, P. (1974). Locomotor movements generated by the deafferented spinal cord. Acta Physiol. Scand. 91, 38A-39A.

Grillner, S. & Zangger, P. (1975). How detailed is the central pattern generation for locomotion? Brain Res. 88, 367-371.

Grillner, S. & Zangger, P. (1979). On the central generation of locomotion in the low spinal cat. Exp. Brain Res. 34,

241-261.

Hulliger, M., Matthews, P.B.C. & Noth, J. (1977a). Static and dynamic fusimotor action on the response of Ia fibres to low-frequency sinusoidal stretching of widely ranging amplitude. J. Physiol. 267, 811-838.

Hulliger, M., Matthews, P.B.C. & Noth, J. (1977b). Effects of combining static and dynamic fusimotor stimulation on the response of the muscle spindle primary ending to sinusoidal stretching. J. Physiol. 267, 839-856.

Hulliger, M. (1984). The mammalian muscle spindle and its central control. Rev. Physiol. Biochem. Pharmacol. 101, 1-110.

Hulliger, M., Zangger, P., Prochazka, A. & Appenteng, K. (1984). Fusimotor "set" vs α-γ linkage in voluntary movement in cat. In Proc. 7th Int. Congr. EMG, ed. Struppler, A. & Weindl, A., pp. 57-64. New York: Springer.

Jami, L. & Petit, J. (1981). Fusimotor actions on the sensitivity of spindle secondary endings. In Muscle Receptors and Movement, ed. Taylor, A. & Prochazka, A., pp. 51-56. London: MacMillan.

Laporte, Y. & Emonet-Denand, F. (1973). Evidence for common innervation of bag and chain muscle fibres in cat spindles. In Control of Posture and Locomotion, ed. Stein, R.B., Pearson, K.G., Smith, R.S. & Redford, J.B., pp. 119-126. Plenum Press: New York.

Loeb, G.E. (1984). The control and responses of mammalian muscle spindles during normally executed motor tasks.

Exercise and Sport Sciences Reviews 12, 157-204.

Loeb, G.E. & Duysens, J. (1979). Activity patterns in individual hindlimb primary and secondary muscle spindle afferents during normal movements in unrestrained cats. J. Neurophysiol. 42, 420-440.

Loeb, G.E. & Hoffer, J.A. (1981). Muscle spindle function during normal and perturbed locomotion in cats. In Muscle Receptors and Movement, ed. Taylor, A. & Prochazka, A., pp. 219-228. London: MacMillan.

Matthews, P.B.C. (1972). Mammalian Muscle Receptors and their Central Actions. London: Arnold.

Matthews, P.B.C. (1973). The advances of the last decade of animal experimentation upon muscle spindles. In New Developments in Electromyography and Clinical Neurophysiology, ed. Desmedt, J.E., pp. 95-125. Basel: Karger.

Matthews, P.B.C. (1981a). Evolving views on the internal operation and functional role of the muscle spindle. J. Physiol. 320, 1-30.

Matthews, P.B.C. (1981b). Muscle spindles: their messages and their fusimotor supply. In Handbook of Physiology. Sect. 1: The Nervous System, vol. II, pt.1, ed. Brooks, V.B., pp. 189-228. Bethesda, MD: Am. Physiol. Soc.

Matthews, P.B.C. & Stein, R.B. (1969). The sensitivity of muscle spindle afferents to small sinusoidal changes of length. J. Physiol. 200, 723-743.

Morgan, D., Prochazka, A. & Proske, U. (1984). Single

dynamic fusimotor neurons can maintain firing of cat soleus Ia afferents during rapid muscle shortening. J. Physiol. 353, 39P.

Murphy, P.R., Stein, R.B. & Taylor, J. (1984). Phasic and tonic modulation of impulse rates in γ-motoneurons during locomotion in premammillary cats. J. Neurophysiol. 52, 228-243.

Orlovsky, G.N. & Shik, M.L. (1976). Control of locomotion: neurophysiological analysis of the cat locomotor system. Int. Rev. Physiol. 10, 281-317.

Patla, A.E., Calvert, T.W. & Stein, R.B. (1985). Model of a pattern generator for locomotion in mammals. Am. J. Physiol. 248, R484-R494.

Pearson, K.G. & Duysens, J. (1976). Function of segmental reflexes in the control of stepping in cockroaches and cats. In Neural Control of Locomotion, ed. Herman, R.M., Grillner, S., Stein, P. & Stuart, D.G., pp. 519-538. New York: Plenum Press.

Perret, C. (1976). Neural control of locomotion in the decorticate cat. In Neural Control of Locomotion, ed. Herman, R.M., Grillner, S., Stein, P. & Stuart, D.G., pp. 587-615. New York: Plenum Press.

Perret, C. & Berthoz, A. (1973). Evidence of static and dynamic fusimotor actions on the spindle response to sinusoidal stretch during locomotor activity in the cat. Exp. Brain Res. 18, 178-188.

Perret, C. & Buser, P. (1972). Static and dynamic fusimotor activity during locomotor movements in the cat. Brain Res.

40, 165-169.

Prochazka, A. (1981). Muscle spindle function during normal movement. Int. Rev. Physiol. 25, 47-90.

Prochazka, A. & Hulliger, M. (1983). Muscle afferent function and its significance for motor control mechanisms during voluntary movements in cat, monkey and man. In Motor Control Mechanisms in Health and Disease, ed. Desmedt, J.E., pp. 93-132. New York: Raven.

Prochazka, A., Westerman, R.A. & Ziccone, S.P. (1976). Discharges of single hindlimb afferents in freely moving cat. J. Neurophysiol. 39, 1090-1104.

Prochazka, A., Westerman, R.A. & Ziccone, S.P. (1977). Ia afferent activity during a variety of voluntary movements in the cat. J. Physiol. 268, 423- 448.

Severin, F.V. (1970). The role of the gamma motor system in the activation of the extensor alpha motor neurones during controlled locomotion. Biofizika 15, 1096-1102.

Severin, F.V., Shik, M.L. & Orlovsky, G.N. (1967). Work of the muscles and single motor neurones during controlled locomotion. Biofizika 12, 660-668.

Shefchyk, S., Stein, R.B. & Jordan, L.M. (1984). Synaptic transmission from muscle afferents during fictive locomotion in the mesencephalic cat. J. Neurophysiol. 51, 986-997.

Shik, M.L. & Orlovsky, G.N. (1976). Neurophysiology of locomotor automatism. Physiol. Rev. 56, 465-501.

Sjostrom, A. & Zangger, P. (1975). α-γ linkage in the spinal

generator for locomotion of the cat. Acta Physiol. Scand. 94, 130-132.

Sjostrom, A. & Zangger, P. (1976). Muscle spindle control during locomotor movements generated by the deafferented spinal cord. Acta Physiol. Scand. 97, 281-291.

Sokolnikoff, I.S. & Redheffer, R.M. (1966). Mathematics of Physics and Modern Engineering. New York: McGraw-Hill.

Stein, R.B., Taylor, J. & Murphy, P.R. (1983). Discharge patterns of dynamic and static gamma motoneurons during locomotion in the premammillary cat and ther implications for control of locomotion. Soc. Neurosci. Abstr. 9, 359.

Wetzel, M.C., Atwater, A.E., Wait, J.V. & Stuart, D.G. (1976). Kinematics of locomotion by cats with a single hindlimb deafferented. J. Neurophysiol. 39, 667-678.

Wetzel, M.C. & Stuart, D.G. (1976). Ensemble characteristics of cat locomotion and its neural control. Prog. Neurobiol. 7, 1-98.